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TECHNICAL DIFFICULTIES LEADING TO FALSE INTERPRETATIONS IN BASAL METABOLISM READINGS*

MISS ESTHER I. WILBRECHT, M.T. (ASCP), *New Ulm, Minnesota*

This subject may seem insignificant and irrelevant to most graduate medical technologists, yet the need of applying the results of studies in this field to the correction of the still existing errors continues to be indicated. For many years there has been a desire on the part of some of the medical profession to furnish by means of a mechanical machine, called a metabolor, the final and conclusive evidence of a patient's basal condition. Many doctors assume that a basal metabolism reading is a mechanically accurate test, hence a correct reading is obtained at all times. Too often the accuracy of the reading is accepted without question, with the result that diagnosis has been made, and prescriptions or surgery resorted to, without corroborating evidence, solely upon these findings. Because such errors have often entailed serious consequences, it is not without interest to investigate these common mistakes made in basal metabolism readings and to suggest methods to overcome such errors.

A review of the literature in this field shows that very little is mentioned concerning possible repeated errors that may exist. Most frequently the difficulties can be traced to the personnel administering the tests, often consisting of lay persons who have only learned the mechanics of the machine, without having essential technical instruction and training.

Teaching Methods

A large percentage of the attendants (meaning technicians, medical technologists, nurses, office helpers, or maybe the doctor himself) never received any instruction beyond the simple

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mechanics of the operation. The agent who sold the machine demonstrates to the attendant the mechanism of the equipment, hands her a pouch containing directions for calculations; and the faithful doctor's assistant goes on from there. Genuine understanding of the process is conspicuous by its absence.

Mechanical Errors

The metabolor is a simple mechanical equipment, yet it tells many stories. A competent technologist must be and is able to interpret at a glance the chart as it unfolds itself. The untrained attendant with little or no technical background presupposes that the needle on the chart is infallible; and although a pulse rate of 48 to 50 with a temperature reading of 97.4F. cannot be reconciled with an oxygen consumption of 24 liters, the machine has spoken and its oracular pronouncement must be accepted. A technically and informationally trained operator will be aware of the scores of gremlins that ride the needle up and down the chart, and she will therefore be alerted to the more common causes of error at least, such as the following which the writer has experienced in her activities.

1 - Soda Lime

The most important mechanical error which is constantly being created is the one of never changing soda lime. A survey made in Southern Minnesota communities shows that many nurses employed in physicians offices or clinics who obtained the basal metabolism readings, produced the most errors. This paper is not a criticism of, or reflection on, the nursing profession; but it is meant as a warning and suggestion, that even though this is a mechanical machine, theory and understanding of all factors entering into the results must be considered.

In our department we have frequently been called to clarify some of the unexplainable results. For example, we are told, "We have a patient who has a pulse rate of 52, Temperature of 97.8, blood pressure of 116/92, and a basal reading of 68. Repeated tests produce similar readings. What is wrong with our machine?"

We check carefully for leakage of tubing, for face or mouth inhaler leaks, and most of all for what we have come to consider the most frequent source of trouble, the old problem of soda lime restoration. Usually questioning reveals the startling fact that perhaps one pound of soda lime has in some dim distant past been placed into chamber of the metabolor and this had never been changed. Then there are other attendants who leave the soda lime in the metabolor where it cakes and the machine begins to rust.

Many inquiries have been made by the writer regarding the life of soda lime, but they have yielded very little. The usual answers to these questions can be summarized in this quotation from a leading chemist. "When a patient breathes hard and fast, and the reading of the chart climbs like a skyrocket, you know the soda lime is dead." This then becomes a crude method of elimination and a trial and error method of knowing when to discard soda lime.

In order to arrive at some definite method of disposal of soda lime, we have made a detailed study in our laboratory regarding the usage and life of soda lime. Accurate record of readings, the date on which a new supply of soda lime had been placed into the metabolor, and most important, the number of tests performed, were recorded. After many and repeated recordings we found that an average of 24 patients may be tested with two readings to a test of 6 minutes each, using a two pound can of soda lime. This will give a leeway of perhaps a few tests which could have been performed, tending to greater accuracy. This method of calculation was resorted to after a long trial and elimination method. We concluded that by the time 24 patients had had basal readings difficulty with breathing was usually encountered by the patient. Invariably when removing the mask the patient would take a deep breath of air. This was indication enough that the soda lime was not performing its duty any more.

2 - Leaky Connections

This is invariably the result of poor housekeeping methods. Moisture gets into the metal connections; soon one has a scum, and before one becomes aware of the situation rust and sediment appear. All metal connections must be washed and cleaned after each test, dried thoroughly, and surfaced with vaseline or beeswax around the connections. These connections must be checked before proceeding with the basal test.

3 - Use of Correct Mask or Mouth Inhaler

This comes under the heading of personal preference. Many attendants prefer the use of the mouth inhaler with the nose clamp attachment. Investigations with both have also revealed some information that might be of interest. We have experimented in our department with patients who willingly submitted to the use of both the inhalers. We are convinced that the face inhaler is the better choice. The patient seems much more at ease and the apprehension and fear created by having the nose clamped disappears, thus assuring a more normal reading. In our experiment with both the face and mouth inhaler it was discovered that there was an occasional increase of oxygen con-

sumption ranging from 1.5 to 2 liters per hour with the mouth inhaler and nose clamp.

Another discovery in our survey revealed that many attendants adjust the mouth inhaler and nose clamp, start the motor for the chart reading then leave the room. About 15 to 20 minutes later they suddenly remember that they have a patient who is having a test in some part of the house and rush to turn off the mechanism. This type of attention to a patient having a metabolism test is unpardonable. The slightest movement of the patient's head will sometimes loosen a good connection. Soon the oxygen escapes from the tubing and the test has to be redone. This is time consuming and expensive.

4 - Loose Clothing

Loosening the clothing while the patient is reclining and during the performance of the test is of utmost importance. Experience has proven that a woman patient, wearing a tight corset, consumes twice or three times the amount of oxygen, the result of inability to be relaxed and to expand the diaphragm. The patient is not relaxed, breathes with difficulty, becomes apprehensive and excited, and the results are questionable.

5 - Quiet Surroundings

It is also important that the patient be placed in a quiet room in the doctor's office, or hospital. The ringing of the telephone immediately disturbs the patient. Anyone doing the test will be amazed to see the needle ride up to the end line during this otherwise unnoticed disturbance. Conversations in adjoining rooms, walking past the room in which the patient is submitting to the test, all should be noted as disturbances.

6 - Preparation of Patient

We, as medical technologists, must remember that the welfare of any patient being examined is of prime importance. The patient is the most important person present. We must remember that these patients are mostly lay people with little or no knowledge of diagnostic procedures. Many of these patients are coming to the office or hospital for the first time in their lives for diagnostic study. The medical technologist holds in her power the reassurance that the patient needs. The attendant's kindly and courteous treatment of the patient is an essential factor in securing an accurate reading. Therefore, it is bad practice to explain in front of the patient that the machine is not functioning properly, "something must be wrong with this metabolor" is not a reassuring remark. The best policy is to remedy the situation with as little fuss as possible. It is also bad policy to give instruc-

tions to junior assistants in the presence of a patient. Comments and instructions regarding the procedure are best carried out of the hearing range of the patient. Patients will quickly sense the presence of a competent attendant by the routine of management. The attendant should be distinct in directions to the patient, and if a word of explanation is necessary in the procedure, it should be given. Remember, uncertainty on the part of the attendant and stumbling over methods of procedure and technic soon raise a doubt which will become difficult to erase from the patient's mind. In other words, instruct the patient before the rest period is begun as to the entire procedure of the test.

Our laboratory has endeavored to check and eliminate as many sources of error in basal metabolism readings as possible. We have succeeded in improving our recordings to a degree which enables us to check our results by repeated tests that corroborate the initial reading to a fairly accurate degree. Accuracy in this field is essential; and every suggestion that improves accuracy is worthy of careful consideration.

SUMMARY PREVENTING CONTINUANCE OF ERRORS

1. Have patient in absolute basal rest.
2. Have mechanical and theoretical knowledge of all phases pertaining to the performance of the test.
3. Have metabolor in excellent working condition.
4. Do not have interference with rest.
5. Keep constant watch of graph and patient.
6. Be able to ascertain slightest deviation from normal.
7. Have patient comfortable as to clothes, covers and pillows.
8. See that dentures do not interfere with fitting of mask.
9. Check constantly on the age limit of soda lime, replenish with fresh soda lime at the earliest indication.
10. If the above rules are observed at all times, you may assume that you did your duty. The basal metabolor is a highly technical piece of equipment and can give the diagnostician invaluable information if handled properly and correctly; but, it can also prove extremely detrimental if an inferior technic or slovenly habits are resorted to when performing the tests.

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3. DuBois, Eugene F. M.D.: Basal Metabolism in Health and Disease.
4. Gradwohl, R. B. H. M.D.: Clinical Laboratory Methods and Diagnosis.

CHEMICAL REACTIONS*

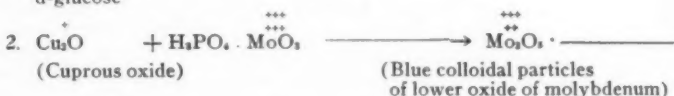
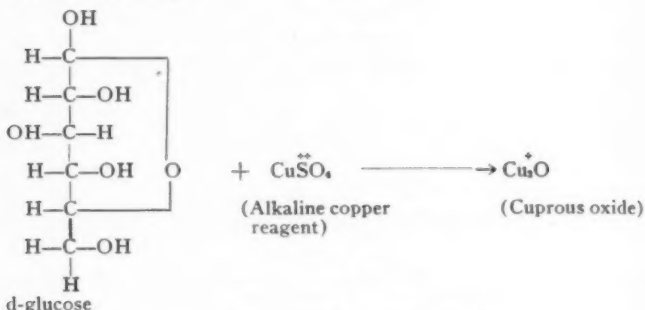
Compiled by

DORIS MAY WALLACE, B.S., M.T., (A.S.C.P.)
Jefferson Davis Hospital, Houston, Texas

The following equations or formulae are those used as teaching aids for students of biochemistry at Jefferson Davis Hospital Laboratory, Houston, Texas. I hope that through these you may also gain a clearer picture of the chemical changes which take place in the various tests.

Glucose Determination (Folin-Wu Method)

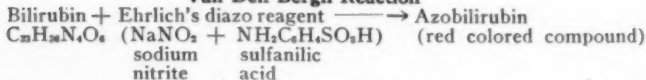
1. Protein free filtrate



Non-Protein Nitrogen (NPN) (Micro-Kjeldahl Method)

1. Protein free filtrate + acid digestion mixture \longrightarrow $(\text{NH}_4)_2\text{SO}_4$
(organic nitrogen) (H_2SO_4) —converts organic nitrogen to inorganic nitrogen
 H_3PO_4 —raises boiling point
 CuSO_4 —catalyst
2. $(\text{NH}_4)_2\text{SO}_4 + \text{HgI}_2 \cdot 2\text{KI} (\text{NaOH}) \longrightarrow$ yellow to brown colloidal particles of ammoniated mercury compound
(Nessler's sol.)

Van Den Bergh Reaction



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Urea Nitrogen

- $$\begin{array}{c} \text{NH}_2 \\ | \\ \text{C} = \text{O} \\ | \\ \text{NH}_2 \end{array} + \text{Urease} + \text{activating phosphate} \longrightarrow (\text{NH}_4)_2\text{CO}_3$$

(enzyme)
- $$(\text{NH}_4)_2\text{CO}_3 + \text{HgI}_2 \cdot 2\text{KI} (\text{NaOH}) \longrightarrow \text{yellow to brown colloidal particles of ammoniated mercury compound}$$

(Nessler's sol.)

Creatin and Creatinine

- $$\begin{array}{c} \text{NH}_2 \\ \diagup \\ \text{HN} \cdot \text{C} \\ \diagdown \\ \text{N} - \text{CH}_2 - \text{COOH} \\ | \\ \text{CH}_3 \end{array} + \text{Heat and pressure to } 120^\circ \text{ C.} \longrightarrow \begin{array}{c} \text{NH} - \text{CO} \\ | \quad \diagup \\ \text{HN} - \text{C} \quad \text{CH}_2 \\ \diagdown \quad | \\ \text{N} - (\text{CH}_3) \end{array}$$

(Creatin) (Creatinine)
- $$\text{Creatinine} + \text{C}_6\text{H}_3\text{OH}(\text{NO}_2)_3 (\text{NaOH}) \longrightarrow \text{Alkaline picrate}$$

$$\begin{array}{c} \text{NO}_2 \\ | \\ \text{H} - \text{C} = \text{C} \\ \diagup \quad \diagdown \\ \text{C} \quad \text{C} \\ \diagdown \quad \diagup \\ \text{C} - \text{C} \end{array} \begin{array}{c} \text{O} \\ || \\ \text{C} - \text{N} - \text{O} - \text{N} = \text{C} \\ || \quad | \quad | \\ \text{O} \quad \text{H} \quad \text{NH} - \text{CO} \\ \quad \quad | \quad | \\ \quad \quad \text{N}(\text{CH}_3) - \text{CH}_2 \end{array}$$

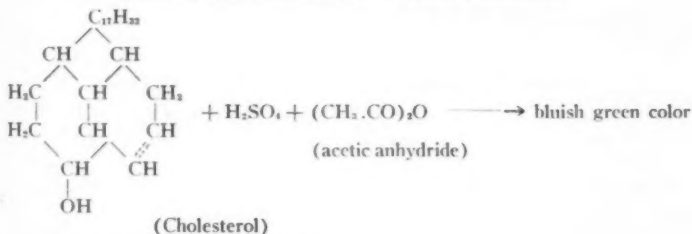
Chloride Determination (Whitehorn Method)

- $$(\text{Cl})^- + \text{AgNO}_3 \longrightarrow \text{AgCl} \downarrow + \text{excess AgNO}_3$$

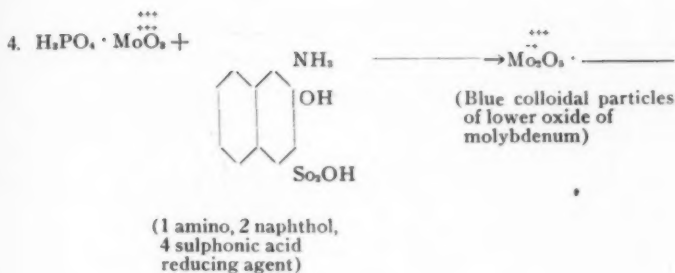
(white ppt.)
- $$\text{AgNO}_3 + \text{NH}_4\text{SCN} \longrightarrow \text{AgSCN} \downarrow \text{ (white ppt.)}$$

(excess from 1) ***
- $$\text{NH}_4\text{SCN} + \text{Fe}_2(\text{SO}_4)_3 \cdot (\text{NH}_4)_2\text{SO}_4 \cdot 2\text{H}_2\text{O} \longrightarrow \text{Fe}(\text{SCN})_3$$

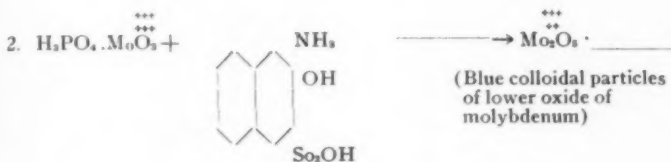
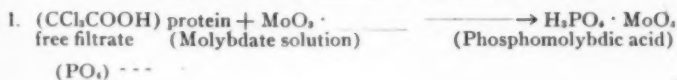
(1st drop in excess of 2) (red solution which gives the salmon pink end point reaction)

Cholesterol (Modification of Bloor's Method)

The above is known as the Lieberman-Burchard reaction.

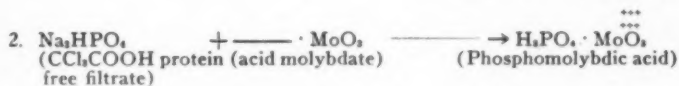
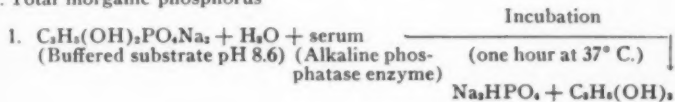


Phosphorus (Briggs' Modification of Bell-Doisy Method)



Alkaline Phosphatase (Bodansky Method)

I. Total inorganic phosphorus



POLIOMYELITIS CONVALESCENT: THROAT FLORA*

SISTER M. ALCUIN, O.S.B., College of St. Scholastica,
Duluth, Minnesota.

The following study was an outgrowth from a finding made in a medical bacteriology class. A blood agar plate inoculated from the throat of a student, a poliomyelitis convalescent, grew only one type of colony. Smears from this culture revealed a Gram-positive coccus in packets. A nutrient broth culture from the same swab demonstrated the identical microorganism. A direct smear from the throat proved to be a pure culture of the same organism. This prompted the identification of the organism. The following laboratory work summarizes the results of the studies we made:

CULTURAL FINDINGS OF THE UNKNOWN ORGANISM

January 17, 1947

January 21, 1947

Nutrient agar	Abundant shiny yellow growth	No significant change in cultural characteristics
Nutrient broth	Slight yellow sediment	
Peptone water	Yellow sediment (indole negative)	
Gelatin	Growth along the stab	
Milk	Coagulation, slightly acid to alkaline	
Potato	Scanty dry yellow growth	
Routine sugars: dextrose, maltose, mannose, sucrose and lactose; no acid or gas.		

The first culture smear confirmed the findings of the direct smear, viz. Gram-positive coccus in packet formation. Culture smear of 1-21-47 showed the same pure culture. The cultural and morphological characteristics answer those attributed to *Sarcina lutea*. Therefore, the unknown organism is *Sarcina lutea*.

Assuming that the student had normal throat flora before her siege of the epidemic poliomyelitis, the question arises how did she acquire this almost sterile throat? Since the normal throat flora^{1,2} are the following: *Leptotrichia buccalis*, the spirochetes: *Treponema macrodentium*, *Treponema microdentium*, *Borrelia buccale*, with their associate, *Fusiformis dentium*, together with the cocci: *Staphylococcus aureus*, *Streptococcus hemolyticus* (in normal throats to the extent of 20 per cent and in winter even a higher percentage), *Streptococcus viridans* (mitior), the harmless parasite, *Neisseria catarrhalis*, the saprophytes (facultative parasites) *Gaffkya tetragena* (tetrads), *Sarcina lutea* (*Staphylococcus teragenus*), and the *Corynebacterium* organism: *C. diphtheriae* (normal in throats of carriers) and *C. pseudodiphthericum*, all unnatural facts associated with the patient's illness need consideration.

The pseudo-sterile throat may be due to possibly one or more of the following causes: 1) Change in acid-base balance of the body fluids of patient; 2) antibiotic action of the virus; 3) the sulfa therapy; 4) the penicillin therapy.

* Read before A.S.M.T. Convention, Denver, Colo., June, 1947.

Bacteria thrive best in an alkaline medium and most of them grow abundantly at a pH of 7.6. This fact prompted the study of the pH of the sputum of the patient and the sputa of healthy classmates. From the table below it becomes evident that the patient consistently had an acid throat:

THE pH OF SPUTA*

pH on Date:	Patient	CONTROL								
		1	2	3	4	5	6	7	8	9
1-24-47	6.5	7.0	7.5	6.8	8.0	8.0	7.2	7.0	8.0	6.5
1-28-47	6.5	6.5	7.4	6.5	7	6.5	6.5	6.5	6.5	6.5

* Normal pH range of sputum 6.0—7.8³.

Projecting the study to the blood stream by doing a carbon dioxide combining power determination on the serum no revealing information was obtained. The reading was 63 volumes per cent. The hydrogen ion concentration may have been toxic to those microorganisms not resistive to drugs to such a degree as to lead to their destruction.

While epidemic poliomyelitis is generally accepted as an infectious disease of viral origin, not too much is known about the causative organism. In an attempt to attain fundamental knowledge about the viruses a Dr. Racker reported in October, 1942 that he obtained from poliomyelitic mouse brains, protein crystals that he thought might be the virus of poliomyelitis and alleged the production in healthy mice in 14 to 72 hours of the acute anterior poliomyelitis.⁴ However in December, 1942 experimenters at the College of Physicians and Surgeons of Columbia University refuted his assertion on the basis that viruses never attain the biometric dimensions equal to the crystals obtained by Racker. Granted that the viruses are crystalline in nature, Racker's crystals were not viruses for they lacked the virulence associated with the latter.⁵ Viruses are known to range from 10 to 250 millimicra in diameter. This is a well-established fact. It is thought that the "polio" virus is a "long thread-like molecule" capable of passing through a membrane with a porosity of 10 millimicra.⁶ There is one other virus that is about the same diameter—these two constitute the two smallest viruses discovered.^{4,7}

It is thought that the virus is contracted either by inhalation sufficient to anchor the organism in the trachea from where it passes through the nerve endings innervating the mucosa of the trachea thus reaching the spinal cord,⁵ or, by mouth, where it grows in the alimentary tract and then passes to the brain.^{7,8} Since, in this case, the virus was not isolated only such evidences

as are associated with the disease justified the establishing of the diagnosis of poliomyelitis. In general the better nourished and more robust member of the family succumbs to the infection.⁹ (The patient in this instance was a well-nourished female weighing 140 pounds, whose blood picture, at this writing, is wbc. 8,400; rbc. 4,200,000; Hb. 12.1 gm.; differential, normal.)

The weather before the time when our patient contracted poliomyelitis was hot and dry, which seems to be the weather peculiar to such epidemics. In a statistical survey of the influence of weather on its incidence, Bowerman reported that acute anterior poliomyelitis may be carried by dust and during hot muggy weather.¹⁰

The first symptom of the oncoming malady in our patient was a severe headache followed by "terrible" emeses. Her throat became exceptionally sore and began to "close" on the third day of illness. The doctor, being called, instituted therapy for a streptococcic sore throat. On the fifth day, the left arm was paralyzed and the patient was put to bed. The following day the paralysis had extended to the muscles of the neck and penicillin treatments were instituted. After two weeks her condition had improved sufficiently to allow visitors, but she was not permitted to get up for a month nor to return to school for three months. During this interim she was assigned to the care of a poliomyelitis therapist and received muscle rehabilitation treatments continuously and was instructed to take them for some time to come. Now, approximately four months since the onset, she has recovered almost completely the use of her arm, but some of her throat muscles "on one side" remain paralyzed. Since this picture answers that of acute anterior poliomyelitis, viruses may have been present in the throat, but nothing confirms their being antibiotic to bacteria.

Relative to the sulfa therapy, a review of the literature has shown that the presence of sulfadiazine in small concentrations in the nasopharyngeal secretions screens out hemolytic streptococci. The bacteriostatic effect of this drug has, however, been overcome by hemolytic streptococci. Further introduction of new strains of this organism into the throat has been prevented by sulfadiazine.¹¹

How concentrated the sulfa drug must be in fluid or soft tissue to be bacteriostatic seems still a matter of speculation. Swift, *et al.*¹², showed that group A hemolytic streptococci were considered susceptible to sulfa drugs in concentrations of 5 milligrams per cent. Hamburger and Lemon¹³ showed that the hemolytic streptococci were promptly and completely removed by "adequate" doses of sulfa drugs, but returned in 5 to 10 days in all but 23 per cent of the throats studied. This would prove in the present study, that were the sulfa dosage adequate, the patient

is either one belonging to the 23 per cent whose throat remained free of streptococci, or the patient's pseudo-sterility must be due to a factor other than the sulfa therapy. Perhaps it is wholly the effect of the penicillin therapy. Spink and collaborators¹⁴ showed that penicillin plus sulfadiazine in treating hemolytic streptococci in tonsillitis, nasopharyngitis, and scarlet fever had no advantage over penicillin alone. Penicillin was, however, not bacteriocidal toward alpha streptococci. Might the clue rest in the fact that all microorganisms were removed by the penicillin therapy save hardy ones, the alpha streptococci in another throat being the counterpart of *Sarcina lutea* in the patient's throat?

If the sulfa therapy were the sterilizing medium one might expect some strains of streptococci to remain among the flora of the patient's throat for Roberg¹⁵ has shown that while some strains of streptococci are susceptible to sulfonamide therapy other streptococci strains are resistant.

There may be no justification for comparing the throat flora in question to the behavior of *Neisseria gonorrhoeae* as reported recently.¹⁶ Culturing the gonococcus in broth reinforced by a bacteriostatic (sulfathiazole) and a duplicate of penicillin and transferred every 48 hours made gonococci acquire an average 700-fold sulfathiazole resistance and only a 182-fold penicillin resistance.

Hirst,¹⁷ in his use of penicillin, showed it up as a bacteriocidal and bacteriostatic substance while acting as an adjuvant in virus research. In his study, sputa from bacterial upper respiratory infections were macerated with equal volumes of nutrient broth, centrifuged at low speed, equal aliquots made, and one treated with 500-1000 oxford units penicillin/100cc., and the other used untreated. These aliquots were inoculated into 13-day old chick embryos. Forty-nine of 55 chicks with untreated inocula died; 47 of 55 chicks with treated inocula lived. The living chicks were found bacterially sterile. Using penicillin thus it was possible to make 33 flu virus and Herpes virus studies from throat washings of virus infected patients. The viruses were not destroyed even with penicillin concentrations of 30,000 units given intramuscularly every three hours and 30,000 to 50,000 units intraspinal.

Rivers¹⁸ asserts that the virus of poliomyelitic is usually found in throat washings and in feces. Scientifically it should be possible to demonstrate, during the acute stages of the infection at least, the virus in the throat even if penicillin and sulfa therapies were instituted, judging from the work of Hirst already quoted.

The only additional attempt to study the action of the chemical factors on the throat flora was to check the sterility of the entire gastro-intestinal tract. For that reason a stool study for flora was made. In comparison with a healthy control it was

apparent that the sterilizing process was limited only to the throat.

Because penicillin is, in the concentration used therapeutically, bacteriostatic rather than bacteriocidal, it seems evident that the drug could not completely sterilize the patient's throat. It is an established fact that penicillin is rapidly excreted by the kidneys¹⁹ and can remain in the blood for 3 hours at the most—therefore it is likely that the throat could not remain sterilized for a prolonged period if we do assume that the drug has the power of such action.

Among the mouth flora that are most susceptible to penicillin are *Staphylococcus aureus*, *Streptococcus viridans*, *Streptococcus hemolyticus* and *Lactobacillus odontolyticus* II. Those insusceptible are one strain of *Staphylococcus albus* and the *Micrococcus albus* species. This fully supports our conclusion because we found a closely related species *Sarcina lutea* which presumably is like the more potent *Staphylococcus*.

Some experimentation has been done on the effectiveness of penicillin biochemically by the various routes of administration and dosage—intramuscularly, intravenously, orally, and otherwise. The poliomyelitis convalescent in this study could not tolerate the drug by mouth. Consequently it was administered intramuscularly. The highest possible concentration attained in her tissues, granted that the drug is accumulative, may have been 0.6 milligrams per cent on weight basis of 140 pounds and 60 per cent of that weight as water. Because the injections were made gluteally the question of how concentrated the penicillin was at the site under observation, viz. the throat, arises. Gerber and his associates²⁰ in his study of the penetration of penicillin into foci of infection (regarding the flora in the throat as infecting organisms limited to a focus) came to the same conclusion as Spink and his collaborators that bacteria vary from strain to strain in their susceptibility to penicillin and that the blood stream "saturation" must be higher for foci of infection other than blood stream infections. Although these workers recognized penicillin as readily diffusible it does not penetrate, via blood stream, the normal spinal fluid, tears, breast milk, and saliva, nor the cornea, lens, brain, nerves, and bone marrow unless huge amounts are administered to attain high blood levels. However they were able to conclude from their studies that some tissues have a selective absorption power on the drug so that concentrations greater than that in the blood may be demonstrated in these tissues. The index or coefficient of resistance they chose arbitrarily to be *Staphylococcus aureus* and setting it as unity showed, among other things, that Actinomycetes are highly resistant. The application to this study of their findings may lie in the possibility of the throat tissues being selectively absorp-

tive of penicillin and that the sterilization process was completed by the drug to the extent of limiting the flora to the one very highly resistant-to-penicillin strain, the *Sarcina lutea*.

Harley, Baty, and Bowie,²¹ in working on the problem of pathogenicity of penicillin-insensitive organisms, showed a resistant strain of *Staphylococcus aureus*, by contact with increased dosages of penicillin, succumbing to the effects of the drug. In our study to evaluate the chemotherapeutic effects of sulfa and penicillin the organism isolated from the patient's throat, *Sarcina lutea*, and two other organisms for controls, were cultured in nutrient broths fortified with varying dilutions of sulfathiazole and penicillin. The control organisms were Gram-positive and Gram-negative organisms representing a coccus and a rod, viz. *Staphylococcus albus* and *A. fecalis*. After 24 hours of incubation at 37° C. subcultures were made with the following findings:

BACTERIOCIDAL EFFECT OF PENICILLIUM AND SULFATHIAZOLE IN VARYING DILUTIONS ON *SARCINA LUTEA*, *STAPHYLOCOCCUS ALBUS*, AND *ALKALIGENES FECALIS*

Penicillium Dilution In %	Growth Present		Sulfathiazole Dilutions	Growth Present	
	24 Hours	48 Hours		24 Hours	48 Hours
0.024	None	None	0.037	None	None
0.0024	None	None	0.0037	All three	All three
0.00024	None	None	0.00037	All three	All three
0.000024	None	None	0.000037	All three	All three
0.0000024	All three	All three	0.0000037	All three	All three

From the invitro tests it is safe to assume that the penicillin was a more effective bacteriocidal agent than the sulfa drug. Furthermore, the tissues were saturated equally as much in the patient under therapy to have had the same destructive effects from penicillin in vivo.

Reviewing studies made on regional injections of penicillin in local infections, Rose and Hurwitz²² have shown that deep injections made locally, i.e. hypodermic injections once daily of 25-50,000 units per cc. (equivalent to 3 milligrams per cent) into furunculotic, cellulitic, abscessed or osteomyelitic areas relieved the pain promptly and spontaneous evacuation of the infection followed.

Schwenlein²³ and co-experimenters showed the effects of penicillin on a patient in a study of the spinal fluid concentration after intravenous injections. They were able to show that the use of penicillin may cause azotemia. Studies on the patient involved in this case made to show the effect of penicillin gave no indications of azotemia at the present writing. This does not exclude a possible temporary azotemia during the interval of chemother-

rapy in this case, but it is conclusive that the convalescent is enjoying comparatively good health.

NITROGEN DETERMINATIONS ON PATIENT

Total Nitrogen Gm. %	N. P. N. Mg. %	Urea Nitrogen Mg. %	A/G Ratio
6.8	24.0	12.0	4.3/2.3

Cholesterol* 178 mg. %

Urinalysis

Color	Cl. amber	Mic.
Reaction	pH 5.5	Wbc.
Sp. Gr.	1.026	6-8 P. H. F.
Alb.	0	Desquam.
Sugar	0	epi. c.
		many

* The cholesterol determination was made to ascertain the degree of glandular involvement proximate to the throat. Because the low, but normal, cholesterol reading it seems safe to say that the sterility of the throat is due to topical features at present. This does not rule out the possibility that the condition was originally initiated by the sulfa and penicillin chemotherapy.

Summary

A poliomyelitis convalescent case is presented in which the throat was sterile except for the presence of *Sarcina lutea*. The possibility of penicillin therapy and sulfa therapy together with or dissociated from antibiotic effects of the causative organism, the virus of poliomyelitis, were considered. It seemed safe to conclude that the sulfa therapy may have inhibited the growth of strains of *Streptococci* and *Staphylococci* normal to the throat. The absence of the spirochetes and bacteria other than the cocci may be due to the combined effects of both penicillin and sulfa therapy. Because of the persistent acid range of the throat secretions or because of possible aerobic conditions as a sequelae to the chemotherapy, the anaerobes were destroyed. Again, might the clue rest in the fact that all microorganisms were removed by the penicillin therapy save the hardy ones, of which alpha streptococci is the classical example, and the alpha streptococci in another throat being the counterpart of *Sarcina lutea* in the patient's throat.

The study of the return of the flora normal to the human throat in the patient is in progress. No clue is given in the literature reviewed as to what may be expected in this case.

We express our thanks to Glenn Denys, M. D., physician, for permitting a study of his patient and to Leo Haanen, Ph. C., for the interpretations of the drug prescriptions. Grateful acknowledgement is made to my students in the Physiological Chemistry, Medical Bacteriology, and Hematology classes for the assistance given in the execution of this study. Special mention is given to:

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SIMPLE PROCEDURE IN THE IDENTIFICATION OF YEAST-LIKE FUNGI*

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Mycology was a known science before the field of bacteriology was discovered. It began in 1677 when Hooke¹ with the aid of a hand-made magnifying lens discovered and described in detail the filamentous organism causing the yellow spots on the Damask rose. The first pathogenic fungi to be discovered and described was the organism causing thrush. The genus had been described by Gmelin² in 1791, but Lagenbeck³ described the pathogenic strain in 1839. Further work was done by Robin⁴ in 1843. About this time the cause of Favus by Schoenlein⁵ and the cause of ringworm by Gruby⁶ were discovered. There was much interest among investigators in mycology until Pasteur and Kock made such startling discoveries in bacteriology. Further investigations were somewhat dwarfed by the important findings in this new field. Attention was again drawn to mycology by Sabouraud⁷ in his scientific work on fungi in 1910.

The global war has emphasized the large number of diseases caused by fungi. It is imperative that the medical technologist be familiar with the technics employed in the isolation and cultivation of fungi. The diagnosis of a fungus infection is made by the physician from the clinical picture. It can be confirmed in the laboratory.

There is a difference between bacteriological technic and mycological technic. The following materials are essential for the study of any fungi:

1. A good microscope.
2. Bottles containing 20% KOH, alcohol, and a fungal stain. (Fungi do not retain the ordinary bacteriological stains. Lacto phenol-cotton blue is very satisfactory.)
3. Sharp teasing needles.
4. Media for the cultivation of the fungi including: Sabouraud's agar for slants and Petri plates; malt agar for the study of mycelium; carrot plugs for the production of asci; hay infusion agar for the study of morphological characteristics; honey agar for the production of giant colonies; and 1% lactose and saccharose broth for sugar fermentations.

Swartz⁸ has classified the pathogenic fungi into 3 groups. The first includes the yeast and yeast-like fungi, the perfect and imperfect. Under the perfect are the Saccharomycetaceae (the true yeasts) and the *Endomyces* (the filamentous yeasts. The imper-

* Given at Tri-State Hospital Assembly, May 6th, 1947, Chicago, Illinois

fect yeast-like fungi includes the *Cryptococcus* (Torula) and *Candida* (Monilia).

In the first group the *Candida* is the most often seen. The organism when found on the mucous membrane causes thrush, vaginitis, and on the skin moniliasis. If found in the lungs, it may cause bronchomoniliasis. The organism may give positive blood pictures with a systemic involvement.

As the yeast-like organisms are frequently found we have employed the procedures for identifying the *Candida* group reported by Burt and Ketchum⁹ in 1941. This is a much less complicated technic than the procedures reported by Martin¹⁰ et al in 1937. The strains are classified by correlating the use of maltose and sucrose for fermentation reactions, the type of mycelium and the time necessary for the presence or absence of asci, the types of giant colonies produced on honey agar in 30 days, and the lesions produced in rabbits following animal inoculation.

When a specimen is received for the identification of a yeast-like organism, the material is streaked on Petri plates of Sabouraud's agar adjusted to a pH 2. The low pH will eliminate much of the secondary contamination. One plate is incubated at 37° C. and the other at room temperature. *Candida* appears as a cream white smooth colony in about 48 hours. It is very similar to *Saccharomyces*, *Endomyces* or Torula.

Some fungi required 10 days or longer for evidence of growth. The organism is transferred to Sabouraud slants for further study. Malt agar plates and carrot plugs are inoculated for study of mycelium and the development of asci. Sugar fermentations are employed. The broth is prepared with Leibig's beef extract, Difco, peptone, NaCl and water. This is sterilized for 20 min. at 15 lbs. pressure. A 10% sugar solution sterilized separately at 10 lbs. pressure for 10 min. is added to the broth in a sufficient amount to make a 1% solution. Brom-thymol-blue is used as an indicator and the pH is adjusted to 7.2. The modified Durham tubes of saccharose and maltose are seeded with 1.5 ml. of a saline suspension from a 48 hour slant of the organism. These are incubated at 37° C. and readings are made in 10 days. Chart No. 1 gives the reactions for each organism. There is much controversy in the literature upon the value of fermentation reactions. In our hands we have had satisfactory results. It is important in preparing the sugar to use pure sugars. We have used Pfanstiehl sugars only. The failure to use pure sugars has resulted in much of the confusion when fermentations were employed.

The presence or absence of mycelium is noted after 48 hours of incubation. The *Saccharomyces* and *Cryptococcus* do not form mycelium. *Endomyces* gives an abundant growth of a fringe-like mycelium. Typical growth is formed by each species of the *Candida* group,

with the exception of *C. parakrusei* which rarely develops any mycelium in 48 hours. The malt agar plates may be studied under the low power objective of the microscope. Certain general characteristics have made it possible to separate the six species when studied in this manner. The various strains of *C. albicans* show clusters of blastospores at the junction of the hypha in from 2-6 days. *C. tropicalis* reveals 2-3 blastospores at the hyphal junction. The general pattern of the growth which extends from the border is of value in differentiating species. *C. tropicalis* produces a fringe-like appearance, *C. pseudotropicalis* is short and rather stubby and there is very little branching. *C. Krusei* has a large amount of growth but can easily be recognized by the spreading type of growth. There is a similarity in the mycelial growth of *C. stellatoidea* and *C. albicans*. However the nakedness of the hypha in cultures of *C. albicans* as compared to a heavier production of blastospores as produced by *C. stellatoidea* aid in the differentiation.

YEAST-LIKE FUNGI

Chart I

Organism	Maltose	Saccharose	Malt Agar Mycellum in 48 Hours	Carrot Plug Asci Formed
<i>C. albicans</i>	AG	A	+	—
<i>C. tropicalis</i>	AG	AG	++	—
<i>C. parakrusei</i>	O	O	—	—
<i>C. krusei</i>	O	O	+	—
<i>C. pseudotropicalis</i>	O	AG	+	—
<i>C. stellatoidea</i>	AG	O	+	—
<i>Endomyces</i>	O	AG	++	+
<i>Blastomyces (Dermatitidis)</i>	O	O	—	+
<i>Saccharomyces</i>	AG	AG	—	+
<i>Cryptococcus (Torula)</i>	O	A	—	—

Carrot plugs are prepared in the same manner as the potato slants. After inoculation they can be incubated at room temperature. The ascospores may develop in the *Saccharomyces* in about 7 days but the slants should be incubated 30 days or longer before considering them negative. *Blastomyces dermatitidis* is included in this chart because it appears in the diseased tissue as a double contoured yeast-like organism and will give no reactions when fermentations are employed.

At the present time we are using the giant colony and animal inoculation for confirmation only. If the organism is thought to be in the *Candida* group and an early report is indicated, animal inoculation may be employed to advantage. From 3.5-6 million organisms per 100 g. body weight are suspended in saline and

given intravenously in a healthy adult rabbit weighing from 1800-2100 grams. If the organism is pathogenic the animal will die in 4-6 days (usually on the 5th day). When the animal is posted the typical picture of large abscessed kidneys is found. These were described by Benham¹¹ in 1931 and Stovall and Pessin¹² in 1933. *C. albicans* is the only organism that is pathogenic for rabbits with this inoculum. *C. tropicalis* may be lethal when a much larger inoculum is employed.

In summarizing, a method has been described for the identification of the yeast-like organism by correlating the fermentation reactions with the production of mycelium on malt agar and the production of asci on carrot plugs.

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SYMPOSIUM: CARDIOLIPIN*

KOLMER CARDIOLIPIN COMPLEMENT-FIXATION TEST

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Introduction: The technique for conducting the Kolmer Quantitative Test using *cardiolipin antigen* is the same as the routine followed in the regular Kolmer Test. The usual 5-dilutions of test serum are made and titrations of hemolysin and complement carried out prior to use. The preparation of antigen (used in the complement titration and in the test proper) is the only new feature. All other reagents employed (saline, hemolysin, sheep cell suspension, etc.), temperatures, and time intervals are the same as specified by the Kolmer method. Absorption of sera with washed packed sheep cells to remove natural anti-sheep hemolysins is an optional step. Here in our laboratory, we absorb routinely.

Procedure: Exactly how do we prepare Kolmer cardiolipin antigen? Solutions of cardiolipin, lecithin, and cholesterol in absolute alcohol are used in the following concentrations:

Cardiolipin	0.03%
Lecithin	0.05%
Cholesterol	0.3%

These concentrations are the results of numerous studies made by Harris and Portnoy using solutions of varied composition and have been thoroughly tested for adequate sensitivity and specificity.

A stock solution of these ingredients in the concentrations stated may be prepared in advance and stored in the dark in a screw-top bottle at room temperature. The screw-top should be lined with tinfoil, and, at no time should the mixture be placed in the refrigerator. The stock solution, in a total volume of 100 cc., and using Lederle products, would be made up as follows: In a 100 cc. volumetric flask, place 15 cc. of 0.2% cardiolipin, 5 cc. of 1% lecithin, and 30 cc. of 1% cholesterol; add absolute alcohol to the 100 mark and mix.

*Editorial Note: On February 29, 1948, the School of Medical Technology of Texas Christian University and Harris Memorial Methodist Hospital of Fort Worth, Texas, offered a one day seminar for registered medical technologists. John J. Andujar, M.D., FACP, Director, and the school staff provided the instructors for the sessions. The morning was devoted to Cardiolipin and the afternoon to Rh-Hr (CDE-cde) factors. The program included demonstrations of all the procedures. This material is herewith presented in order that the membership of the American Society of Medical Technologists may share the instruction which those representatives of the Texas, Arkansas, Oklahoma, and Louisiana Societies in attendance received.

The antigen emulsion is made up in a final dilution of 1:150 in Kolmer saline. For example, 0.5 cc. of the stock mixture is added slowly, by dropping, to 74.5 cc. of saline. The diluted antigen is now ready to add to the complement titration and should be stored in the refrigerator until the test sera have been properly diluted and the units of hemolysin and complement determined.

The test proper, following the Kolmer quantitative method should include the five dilutions of serum plus a serum control, and also controls using positive and negative serum; not to mention the hemolysin, antigen, and sheep cell suspension controls necessary for accurate interpretation of results. Repeat hemolysin titrations after overnight incubation to determine the degree of complement deterioration should also be carried out.

Data: A large number of sera were tested using regular Kolmer antigen and cardiolipin antigen in parallel series. The following results were noted:

TESTS ON BLOOD OF KNOWN SYPHILITIC STATUS

Syphilitic Status	Antigen	Results			
		Negative	Doubtful	Positive	Total Tests
Primary	Regular Cardiolipin	59	3	11
		49	7	18
Secondary	Regular Cardiolipin	31	8	66
		21	12	78
Latent	Regular Cardiolipin	173	48	2008
		91	57	2064
Tertiary	Regular Cardiolipin	71	12	539
		31	7	585
Congenital	Regular Cardiolipin	12	4	33
		8	3	37
Doubtful or unknown	Regular Cardiolipin	732	26	28
		698	47	36
Nonsyphilitic (?)	Regular Cardiolipin	363	2	4
		359	3	7
Totals	Regular Cardiolipin	1441	103	2689	4233
		1257	136	2825	4218

Discussion: The use of regular Kolmer antigen and cardiolipin antigen in parallel series has shown a definite superiority in specificity and sensitivity with the cardiolipin antigen.

The parallel series of complement titrations and test runs using

RESULTS ON FALSE-POSITIVES

No. of Cases	Diagnosis	Results			
		Antigen	Negative	Doubtful	Positive
2	Acute infectious lymphocytosis	Regular Cardiolipin	1 1	0 1	1 0
14	Acute infectious monocyctosis	Regular Cardiolipin	5 8	5 4	4 2
48	Acute upper respiratory infection	Regular Cardiolipin	35 41	4 3	9 4
31	Malaria, tertian, early	Regular Cardiolipin	19 25	8 3	4 3
37	Pregnancy, 2d, 3, trim.	Regular Cardiolipin	30 34	2 1	5 2
11	Puerperium, early	Regular Cardiolipin	6 9	3 0	2 2
7	Brucellosis	Regular Cardiolipin	4 3	2 2	1 2
8	Vaccinia (youths)	Regular Cardiolipin	3 7	3 0	2 1
5	Viral pneumonia	Regular Cardiolipin	1 3	1 1	3 1
17	Vincent's angina	Regular Cardiolipin	12 10	1 3	4 4
180	Totals	Regular Cardiolipin	116 141	29 18	35 21

both antigens yielded some interesting observations. Fewer anticomplementary reactions were encountered with cardiolipin antigen than with regular antigen. The percentage, though small, is worth mentioning.

In most instances, the complement titrations using both antigens yielded the same readings. A series of studies made by Mrs. Shaw of San Antonio has shown that a higher unit of complement frequently resulted in the cardiolipin complement titration than in the regular complement titration; and that when parallel titrations yielded the same *unit*, supplementary hemolysin titrations following overnight incubation permitted the use of higher dilutions of hemolysin in the cardiolipin series than in the regular series. Our tests have borne this out. These increased dilutions of complement and hemolysin may account for

the increased sensitivity of the cardiolipin complement-fixation test.

Our results with Kolmer testing of spinal fluid using both antigens have been comparable to the serum studies made.

Conclusion: Cardiolipin antigen is reproducible and stable. Satisfactory results are obtainable many days after preparation of the antigen mixture. By comparison, we all know that different lots of regular Kolmer antigen vary considerably in specificity and sensitivity. Reproducibility and stability, even after prolonged storage of solutions, are certainly desirable features. The reproducibility of results between laboratories would surely be a boon to the medical technologist as well as to the clinician who is so often confronted with conflicting reports from laboratories making tests on identical samples of sera.

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THE VDRL TEST

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With the advent of cardiolipin as an antigenic constituent, there were evolved many new flocculation technics. Some of these simply substituted cardiolipin antigen in well-known tests such as the Kline and the Kahn. Others were entirely new.

Probably one of the best known of these new procedures is the VDRL Test. Developed by Harris, Rosenberg, and Reidel of the United States Public Health Service, it is named after the Venereal Disease Research Laboratory at Staten Island, New York. This test is a rapidly performed, simplified technic, with an acceptable degree of sensitivity and specificity. Reagents used are easily standardized and results obtained with the antigen suspension are reproducible in other laboratories. These characteristics make it possible for small laboratories as well as large ones to run the VDRL with ease. Those of you who have run the Mazzini will recognize immediately the similarity between the VDRL and the Mazzini test.

Preparation of the Antigen: Antigen for this test contains 0.03% cardiolipin, 0.27% purified lecithin, and 0.9% cholesterol in absolute alcohol. Each lot of antigen should be serologically standardized by proper comparison with an antigen of known reactivity.

Antigen is dispensed in screw-cap bottles and stored at room temperature in the dark. Tops of the bottles must be lined with tinfoil. Any precipitate noted will indicate changes due to factors such as evaporation or materials introduced by pipettes. If the antigen should contain a precipitate, it should be discarded.

Preparation of Saline: A 1% saline solution is prepared and buffered by adding 0.093 gm. of secondary sodium phosphate and 0.17 gm. of primary potassium phosphate to a liter of formalinized saline. The final pH of this solution is 6.0. It is stored in screw-cap or glass stoppered bottles.

Preparation of Serum: Serum is prepared in the same way as for the Kline Test; that is, heated for 30 minutes at 56° C. Sera to be tested more than four hours after being heated should be reheated at 56° C. for 10 minutes.

Slides: Glass slides with paraffin rings as described by Kline are recommended for use by the authors in the Test Proper. We have found slides with concavities quite satisfactory, however.

Preparation of Antigen Emulsion: 0.4 cc. of buffered saline is pipetted to the bottom of a 30 cc. round, glass-stoppered bottle. Add 0.5 cc. of antigen to the saline, drop by drop (rapidly without long intervals between drops) from the lower portion of a 1 cc. pipette which is graduated to the tip. The bottle should be continuously but gently rotated on a flat surface while adding the antigen. The pipette tip should remain in the upper third of the bottle and rotation should not be vigorous enough to splash saline on to the pipette tip. The last drop of antigen is blown from the pipette without touching it to the saline and the bottle is rotated for 10 more seconds.

Add 4.1 cc. of buffered saline from a 5.0 cc. pipette; the top is placed on the bottle and the bottle is shaken vigorously for 10 seconds (throwing the liquid from top to bottom of the bottle). The antigen emulsion is then ready for use and may be used during one day. One batch of antigen is sufficient for about 250 serum tests. Twice this amount of antigen emulsion may be prepared at one time in a 30 cc. bottle using doubled quantities of antigen and saline. If larger quantities of antigen emulsion are required, more than one mixture should be prepared.

Checking Antigen Emulsion: Each preparation of antigen emulsion should first be checked by testing with known positive and negative sera. This is done by adding one drop of antigen emulsion (1/60 cc.) to 0.05 cc. of each serum and completing the test as described under Qualitative VDRL Test. These tests should present typical positive and negative results.

The antigen emulsion should be dispensed from a 23 gauge, long bevel, hypodermic needle attached to a 1.0 or 2.0 cc.

syringe which may stand in the antigen emulsion bottle when not in use. Approximately sixty drops should be obtained from 1 cc. of antigen emulsion. This can be accomplished by holding the syringe so that the needle bevel is down and the dropping surface horizontal. Increasing the angle at which the syringe is held diminishes the dropping surface and consequently decreases drop size. When allowed to stand, antigen emulsion should be gently mixed before use by rotating the bottle and filling and emptying the syringe.

Qualitative VDRL Test:

1. Pipette 0.05 cc. of heated serum into a ring of a paraffin-ringed glass slide.
2. One drop (approx. 1/60 cc.) of antigen emulsion is then added to each serum.
3. The slides are rotated for four minutes, either by hand on a flat surface or by a Boerner type rotator as used for Klines.
4. Tests are then read immediately after rotation.

Reading and Reporting Results: Tests are read microscopically with low power objective at 100X magnification. The antigen particles appear as short rod forms at this magnitude. Aggregation of these particles into large or small clumps is interpreted as various degrees of positivity.

Reading

No clumping, very slight roughness.
Small clumps.
Medium and large clumps.

Report

Negative (N).
Weakly positive (WP).
Positive (P).

Proper reading and interpretation of test results requires that the technologists have had training and experience in each technique employed, but rough equivalents may be used for describing similarities between the reading of this and other slide tests. The WEAKLY POSITIVE range of this test includes clumpings similar to those reported as plus-minus or one plus, and the POSITIVE range those reported as 2 plus, 3 plus, or 4 plus in other slide tests. However, Harris and associates recommend that these numerals not be used for reporting the results of this test.

Zone reactions, due to an excess of reagin, are recognized by irregular clumping and the loosely bound characteristics of the clumps. The usual positive reaction is characterized by large or small clumps of fairly uniform size and experience will allow differentiation to be made between this type of reaction and the zone reaction wherein large and/or small clumps may be intermingled with free antigen particles.

Whenever a zone type of reaction is suspected, the serum in question should be diluted 1:5 and 1:25 and retested. The maximum reaction produced by either of these dilutions, if greater

than that obtained with undiluted serum, is reported as the test finding.

Data: During a period of four months, from November, 1947, through February, 1948, this laboratory ran parallel tests on 2,532 sera with the Kline Diagnostic (using regular antigen) and the VDRL. Kline Exclusion Tests were run on 83 of these sera.

- (1) 2,511 were in complete agreement yielding 2,471 completely negative results and 40 positive results confirmed by positive Kolmers (both regular and cardiolipin).
- (2) 4 sera gave positive results with the Kline Diagnostic and the VDRL but had negative Kolmers. Four other sera gave positive results with both the Kline and the VDRL and had anticomplementary Kolmers.
- (3) 8 were in complete disagreement; 5 were in partial disagreement. The majority of these last 13 sera gave negative results with the Kline Diagnostic antigen and Kolmer antigen, and positive or doubtful results with the VDRL.

An analysis of the small number of sera which gave any positive reaction whatsoever would seem to indicate that the VDRL is more sensitive than the Kline Diagnostic and slightly less specific. A study of the sera run with Kline Exclusion antigen in addition to the VDRL and Kline Diagnostic, shows that the VDRL is definitely more specific than the Kline Exclusion and has a sensitivity approaching that of the Kline Exclusion.

Summary: The VDRL test is a rapidly performed microflocculation test with an acceptable degree of sensitivity and specificity. Antigen emulsion is easily and rapidly prepared with a minimum amount of manipulation involved. Only two reagents are needed and both are stable (antigen and buffered saline). Reading of results may be correlated to those of other slide tests, thus making it possible for technologists to run the VDRL Test without training first under the author serologists.

REFERENCE:

- Harris, A., Rosenberg, A. A., and Reidel, L. M.: A microflocculation test for syphilis using cardiolipin. *J. Ven. Dis. Inform.* 27: 1946.

NOTES ON THE VDRL TEST

Source of Antigen: Lederle Laboratories at present markets the Cardiolipin and Lecithin solutions separately. For details, consult our release on the Kline Test.

Preparation of Antigen: Antigen for this test contains 0.03% cardiolipin, 0.27% purified lecithin, and 0.9% cholesterol in absolute alcohol. Each lot of antigen should be serologically standardized by proper comparison with an antigen of known reactivity.

Antigen is dispensed in screw-cap bottles and stored at room temperature in the dark. Tops of the bottles must be lined with tinfoil. Any precipitate noted in the antigen will indicate changes due to factors such as evaporation

or materials introduced by pipettes. If the antigen should contain a precipitate, it should be discarded.

At present, Lederle's stock solution of Cardiolipin is 0.2% (2 mg./cc.) and the Lecithin sold is 1% (10 mg./cc.)

To dilute these reagents for proper concentration, use the formula:

mg. in 1 cc. of stock solution : 1 cc. :: mg. needed to prepare desired amount of given antigenic mixture : (x) cc.

To find the amount of each stock solution needed to prepare 100 cc. of antigen containing 0.03% cardiolipin (30 mg./100 cc.), 0.27% lecithin (270 mg./100 cc.), and 0.9% cholesterol (900 mg./100 cc.), set up equations as follows:

2:1 :: 30:x

x = 15 cc. of stock solution of cardiolipin

10:1 :: 270:x

x = 27 cc. of stock solution of lecithin

Weigh out 900 mg. of cholesterol (C. P. Pfanstiehl). Place the correct amounts of stock solutions of cardiolipin and lecithin in a 100 cc. volumetric flask and add the cholesterol. Add absolute ethyl alcohol to the 100 mark but allow time for the cholesterol to dissolve completely before making up to this total volume.

Preparation of Saline:

Formaldehyde, neutral, reagent grade.....	0.5 cc.
Secondary sodium phosphate ($\text{Na}_2\text{HPO}_4 \cdot 12 \text{H}_2\text{O}$)...	0.093 gm.
Primary potassium phosphate (KH_2PO_4)	0.170 gm.
Sodium chloride, A.C.S.....	10. gm.
Distilled water	1000. cc.

Preparation of Serum: Serum is prepared in the same way as for the Kline Test, that is, heated for 30 minutes at 56° C. Serums to be tested more than four hours after being heated, should be reheated at 56° C. for 10 minutes.

Preparation of Slides: Glass slides (2 x 3 in.), with 12 paraffin rings of approximately 15 mm. inside diameter as described by Kline, are used. We have used agglutination slides with 12 concavities successfully.

Preparation of Antigen Emulsion:

1. Pipette 0.4 cc. of buffered saline to the bottom of a 30 cc., round glass stoppered bottle.
2. Add 0.5 cc. antigen, drop by drop (rapidly, without long interval between drops) from the lower portion of a 1.0 cc. pipette graduated to the tip, directly into the saline while continuously but gently rotating the bottle on a flat surface. Keep pipette tip in upper third of the bottle. Blow the last drop of antigen from the pipette without touching it to the saline.
3. Rotate bottle for 10 more seconds.
4. Add 4.1 cc. of buffered saline from a 5 cc. pipette. Place top on bottle and shake vigorously for 10 seconds.

The antigen emulsion is then ready for use and may be used during one day. During the summer months when the temperature is high and humidity is low, antigen emulsion should be stored in the refrigerator.

Checking Antigen Emulsion: Test antigen emulsion with known positive and negative serums. This is done by adding one drop of antigen emulsion to 0.05 cc. of each serum and completing the test described under Qualitative VDRL. The results should present typical positive and negative serums.

Qualitative VDRL Test:

1. Pipette 0.05 cc. heated serum into one ring of a glass slide.

2. Add one drop (approximately 1/60 cc.) antigen emulsion onto each serum. Antigen emulsion should be dispensed from a 23 gauge, long bevel, hypodermic needle attached to a 1.0 or 2.0 cc. syringe which is allowed to stand in the antigen emulsion bottle when not in use. Approximately 60 drops should be obtained from one cc. of antigen emulsion.
3. Rotate slides for four minutes. Automatic rotator set at 180 r.p.m. may be used. If rotated by hand on a flat surface, movement should circumscribe a two-inch circle 120 times per minute.
4. Read tests immediately after rotation.

Reading and Reporting Test Results: Tests are read microscopically at 100x magnification. The antigen particles appear as short rod forms at this magnification. Aggregation of these particles indicates various degrees of positivity.

Reading	Report	Similar to Kline Reading of:
No clumping	Negative (N)	Negative
Very slight roughness	Weakly positive (WP)	Plus-minus, one plus
Small clumps	Positive (P)	Two, three and four plus
Medium and large clumps		

If zone reaction is suspected, characterized by irregular clumping and loosely bound clumps, dilute serum with saline 1:5 and 1:25. Repeat test. If the maximum reaction produced by either of these dilutions is greater than that obtained with undiluted serum, report as the test finding.

NOTES ON KLINE CARDIOLIPIN TESTS

(As of February 29, 1948)

Antigen, Source: Cardiolipin-Lecithin Kline antigen may be purchased ready mixed from LaMotte Chemical Products Company, Baltimore, Maryland. When stored in a cool, dark place the 5 cc. vials will keep for at least six months. The two components may be purchased as separate packages from Lederle Laboratories, New York, with individual bottles having 0.2% of Cardiolipin and 1% of Lecithin in absolute alcohol. The cost of ready mixed antigen is approximately the same as regular Kline antigen. The LaMotte mixture contains ten parts of Lecithin to 1 of Cardiolipin, an excellent or so-called "optimal" proportion. More lecithin raises the sensitivity; less lecithin will drop even the specificity.

Antigen, Calculation: The LaMotte antigen is, of course, ready to use and can be made directly into the emulsion. If the separate (Lederle) Cardiolipin and Lecithin are used, the exact weight of cardiolipin should be found on the label and the same is true of lecithin. Thus, the .2% cardiolipin actually may have 1.98 mg. per cc. and the 1% lecithin actually may have 10.4 mg. per cc. rather than exactly 10 mg. The formula for mixing 10 parts of lecithin to 1 of cardiolipin in such an instance would be as follows:

$$\frac{1.986 \times 10}{10.4} = 1.978 \text{ cc. lecithin to each cc. of cardiolipin.}$$

While our work was done with a Lederle solution of 3% lecithin, the weaker solution can be vacuum distilled down from its marketed 1% to either 2 or 3%.

Emulsion Formula: 0.85 cc. of distilled water (pH about 6); 1.0 cc. of 1 per cent cholesterol solution (1 gram Pfanstiehl cholesterol C. P., ash free, precipitated from alcohol in 100 cc. absolute alcohol); 0.1 cc. of Lecithin-Cardiolipin antigen as prepared above; 2.45 cc. of 0.85 per cent sodium chloride (C.P. or reagent, Merck) solution (pH about 6).

Emulsion Technique: The technique of preparing the above emulsion is as follows:

- (1) Into a glass stoppered 1-ounce bottle pipette the 0.85 cc. of distilled water.
- (2) Holding the bottle at an angle in the right hand, pipetting with the left, run the 1 cc. of 1 per cent cholesterol solution SLOWLY along the side of the neck of the bottle (15 seconds for 1 cc.). During the entire 15 seconds the container is shaken or rotated fairly vigorously.
- (3) The bottle is then gently rotated on a flat surface for 20 seconds.
- (4) Holding the bottle at an angle again, the 0.1 cc. of Cardiolipin-lecithin antigen is pipetted against the side of the neck of the bottle.
- (5) The bottle is stoppered and shaken vigorously (the fluid thrown from bottom to stopper and back) for one minute. (In shaking the bottle vigorously after addition of the antigen, it is not necessary to strike the bottle itself against the free hand. No less vigorous shaking can be secured by striking hand against hand.)
- (6) Lastly, the 2.45 cc. of 0.85 per cent sodium chloride is allowed to run in quite rapidly, the bottle is stoppered again and shaken less vigorously than previously for 30 seconds.

The emulsion when examined under the microscope at a magnification of about 100 times, shows numerous very fine discrete particles, but no clumps whatever.

Kline Cardiolipin Diagnostic: (Kline Standard) (Uncentrifuged Emulsion): The emulsion as prepared is ready for use. IT IS NOT HEATED as is the emulsion with regular Kline antigen.

Kline Cardiolipin Exclusion (Centrifuged Emulsion): Pipette $2\frac{1}{2}$ cc. of unheated emulsion into a round bottom tube and centrifuge for 8 minutes at about 1450 R.P.M. The time and speed of centrifugation are of the utmost importance and may vary in different laboratories. Several trials will show the most suitable time and speed. Decant the supernatant fluid, add from 1 to 1.3 cc. saline, shake fairly vigorously (15 seconds) to completely and uniformly distribute the particles.

Test Proper:

- (1) Pipette 0.05 ml. heated serum into a ring of a paraffin-ringed glass slide (glass slide with concavities may be used).
- (2) Add one drop of Kline antigen emulsion (about 0.008 cc.) to each serum.
- (3) Rotate for four minutes. (Automatic rotator set at 190 r.p.m. may be used. If rotated by hand on a flat surface, the slide should circumscribe a circle $\frac{3}{4}$ inch in diameter 190 times per minute.)
- (4) The results are examined at once through the microscope at a magnification of about 120 times with the light cut done as for the study of urinary sediments and reported in terms of pluses according to the degree of clumping and the size of the clumps.

Note: Dr. Kline now proposes to discontinue the Kline Exclusion and Kline Diagnostic Tests with regular antigens. He will sponsor only the Cardiolipin version of the Kline Diagnostic using an entirely new title, "THE KLINE STANDARD TEST."

SYMPOSIUM: Rh-Hr AGGLUTININS

TESTS FOR ANTI Rh-Hr ANTIBODIES

BARBARA BICKLE, MT (ASCP), *Instructor in Hematology*

The phase of the Rh (DCE) factors with which we are usually most vitally concerned is the role it plays in erythroblastosis fetalis and in transfusion reactions. A small portion of the population will, with continued or repeated stimulation, develop antibodies against one or more of the Rh-Hr factors. The commonest type of sensitization is against the Rh_o or D factor. The rarity of any sensitization at all is due to the fact that only about 1 in every 40 or 50 individuals is readily sensitized to the Rh_o factor; and sensitization to any of the other factors, which some claim are probably less antigenic, is even rarer. But it is with this group of readily sensitized individuals that we are concerned, and especially with the determination of their sensitization during and after pregnancy and in repeated blood transfusions.

There are believed to be three types of antibodies which cause reactions to the Rh factors. It must be emphasized that, while the majority of these antibodies cause agglutination *in vitro*, their action *in vivo* is hemolytic. The Rh agglutinin is a warm agglutinin. The highest titre is generally found 8 to 20 days after delivery. Antibodies from a previous pregnancy may persist even during pregnancy with an rh negative fetus.

The clinical application of these antibody titrations is mainly during the pregnancy of an rh negative woman and in repeated blood transfusions to an rh negative person. In the case of pregnancy, the patient is carefully watched for a rise in antibody titre. When such a rise is pronounced, it is sometimes taken as an indication for early induced labor or some such measure. This is, of course, a decision for the physician to make. Because of the extra handicap to the child such a means is usually taken only in the case of a definite rise in titre when the danger of an erythroblastotic infant, a macerated fetus, or other results of sensitization seem great.

In the case of repeated blood transfusions, any patient showing any antibody titre should receive only rh negative blood, since the administration of positive blood may result in a severe hemolytic reaction or death.

Only a very small amount of Rh positive cells are necessary for an easily sensitized woman to build up antibodies and pregnancy offers two conditions most favorable for antibody production—slow administration of the antigen and action of the antigen over a long period of time.

The first test is for the saline agglutinins, which are the first

type of antibody. This type of agglutinin was the first to be discovered and for a while was thought to be the only type of antibody for these factors. The patient's serum is diluted with saline to concentrations of 1:2, 1:4, 1:8, 1:16 and higher dilutions as necessary, remembering that when the number of dilutions is kept down, the cost of the test is also lower. The antigen, cells of Group O, and Type Rh₁, Rh₂, and rh, must first be washed with saline. Three rows of five Kahn type test tubes are set up. One drop of the patient's undiluted serum is put in the first tube of each row, one drop of 1:2 in the second, 1:4 in the third, etc. To all tubes in the first row is added one drop of a 2% suspension of Rh₁ cells in saline, Rh₂ cells are added to the second row, and rh cells to the third row. Shake to mix and incubate for 60 minutes at 37° C. The tubes are read microscopically and macroscopically for agglutination. Absence of agglutination in all tubes constitutes a negative test; a positive test is reported as positive to "X" titre, "X" being the highest dilution of the patient's serum which still shows agglutination. If there is agglutination with only Rh₁ cells, the antibody is of specificity Rh' or anti-C; agglutination with only Rh₂ cells indicates an antibody of specificity Rh'' or anti-E. If both Rh₁ and Rh₂ cells are agglutinated, the antibody may be of specificity Rh₁, Rh₂, or Rh₀. To differentiate these agglutinins, cells of the rare groups Rh', Rh'', and Rh₀ are necessary.

The first indications of the existence of antibodies other than the saline agglutinins, were observed in serums demonstrating the prozone phenomenon and in patients who had obviously been sensitized but showed only a very low titre or no agglutination at all with this test. The second order of antibodies was found, which will not usually react in saline but will in albumin or plasma, or will give a much higher titre in albumin or plasma than in saline.

When the saline test is negative, the test for blocking antibodies may be carried out on the same tubes. One drop of anti-Rh₀ serum of known potency is added to each tube. They are then reincubated for one hour at 37° C. and read microscopically for agglutination. Strong agglutination in all tubes is a negative test. Absence of agglutination in some or all tubes, or weak agglutination, is a positive test. The titre is the highest dilution of the patient's serum in which agglutination is inhibited.

The third test for antibodies is the conglutination or albumin test. This is carried out as the saline test, except that all dilutions of cells or serum are made in 20% bovine albumin or type AB plasma. Otherwise the technique and reading of the test is the same as that for the saline agglutinins.

The fourth test is known variously as the Coombs or the

"developing test." The antibodies, which are the third order of antibodies, are capable of attacking the red cell but show no visible agglutination with the previous tests. This antibody is sometimes called cryptagglutinin. The diluted sera are mixed with the test cells as before. When no agglutination appears, the cells are washed with saline, centrifuged and the supernatant fluid removed three times. Then, one drop of anti-human globulin serum is added. They are then reincubated, centrifuged slowly and read microscopically and macroscopically for agglutination. Agglutination indicates the presence of a globulin on the red cells which cannot be removed by thorough washing, or the cryptagglutinin. They are reported the same as in the previous tests.

This serum has also been found very helpful as a diagnostic test for erythroblastosis fetalis, even in sub-clinical cases. For this, a weak suspension of the infant's cells, taken from the cord or within 24 hours after birth, are washed with saline three times and a drop of the anti-human globulin serum added. They are read macroscopically and microscopically for agglutination after incubation for 60 minutes and slow centrifugation. Agglutination indicates the presence of the disease even in the absence of clinical symptoms.

As you can see, the testing for anti-Rh agglutinins has progressed from the relatively simple saline agglutination to agglutinins more complex and more difficult to demonstrate. And, as these different agglutinins, and the rarer agglutinins for the factors others than Rh₀ or D have been demonstrated, the etiology of more and more cases of erythroblastosis fetalis has been found. The main applications of these tests is during pregnancy of an Rh negative woman suspected or known to have been sensitized, and in determining the prognosis of the pregnancy, and in the prevention of transfusion reactions in the case of repeated transfusions to Rh negative individuals. It must also be emphasized that these tests are of equal importance with those patients who are suspected or known to be sensitized to factors other than D.

TEST FOR Rh AGGLUTININS

Principle: The serum of the patient is tested against a saline suspension of red blood cells known to contain the three Rh or CDE factors. Agglutination shows the presence of agglutinins for the corresponding factors.

Procedure: Set up a rack with three rows of Kahn type test tubes, six tubes in each row. In the first tube of each row, put one drop of undiluted patient's serum. In the second tube of each row, put one drop of a 1:2 saline dilution of serum; in the third tube, a 1:4 dilution of serum; in the 4th, a 1:8 dilution; in the 5th, a 1:16 dilution; and in the 6th, a 1:32 dilution of serum. Save the remainder of the last dilution, 1:32, in case further dilutions are necessary. To all tubes in the first row, add one drop of a 2% suspension in saline of Rh₁ cells. To all tubes in the second row, add one drop of Rh₂ cells;

and to all tubes in the third row, add one drop of rh negative cells. All these cells must also be of Group O. Shake and incubate at 37° C. for one hour. Centrifuge at 1,000 rpm for one minute and read macroscopically and microscopically for agglutination.

No clumping in any of the tubes: **NEGATIVE**.

Clumping: **POSITIVE** to "x" titre.

Specificity of the agglutinin:

Clumping against only Rh₁ cells: anti-Rh' or anti-C.

Clumping against only Rh₂ cells: anti-Rh" or anti-D.

Clumping against both Rh₁ and Rh₂ cells: may be anti-Rh₀ or anti-D or, anti-Rh, or anti-CD or, anti-Rh₃ or anti-DE.

To differentiate between the agglutinins in the last case, cells of the rarer types Rh₀, Rh', and Rh" must be obtained.

SCHEME OF BRh GROUPS

Old (Wiener)	New (Fisher-Race)	Frequency (White Population)
Rh ₀	cDe	3.05%
Rh ₁ (Rh ₀ ').....	CDe	50.2
Rh ₂ (Rh ₀ ").....	cDE	15.0
Rh ₁ Rh ₂ (Rh ₀ ' Rh ₀ ")	CDE	11.2
rh'rh".....	CdE	0.01
rh'.....	Cde	1.0
rh".....	cdE	0.5
rh negative.....	cde	14.5

CONGLUTINATION

Principle: The antibody found with the conglutination or albumin test will not, as a rule, act in a crystalline solution. Therefore, all dilutions with this test are made in 20% bovine albumin or AB plasma.

Procedure: Set up a rack with three rows of Kahn type test tubes, six tubes in each row. In the first tube of each row, put one drop of undiluted patient's serum. In the second tube of each row, put one drop of a 1:2 dilution of the serum; in the third tube, a 1:4 dilution of serum; in the 4th, a 1:8 dilution; in the 5th, a 1:16 dilution; and in the 6th, a 1:32 dilution of serum. Save the remainder of the 1:32 dilution of serum in case further dilutions are necessary. To all tubes in the first row, add one drop of a 2% suspension in albumin or plasma, of Rh₁ cells. To all tubes in the second row, add one drop of Rh₂ cells; and to all tubes in the third row, add one drop of rh negative cells. Shake and incubate at 37° C. for one hour. Centrifuge at 1,000 rpm for one minute and read macroscopically and microscopically for clumping.

No clumping in any of the tubes: **NEGATIVE**.

Clumping: **POSITIVE** to "x" titre.

Specificity of the antibody:

Clumping against only Rh₁ cells: anti-Rh' or anti-C.

Clumping against only Rh₂ cells: anti-Rh" or anti-E.

Clumping against Rh₁ and Rh₂ cells: may be anti-Rh₀ or anti-D or, anti-Rh, or anti-CD or, anti-Rh₃ or anti-DE.

To differentiate between the antibodies in the last case, cells of the rarer types Rh_s, Rh', and Rh'' must be obtained.

BLOCKING ANTIBODIES

Principle: The blocking antibodies are antibodies which attack the red cell but do not give any visible reaction.

Procedure: When the saline agglutination test is negative, one drop of anti-Rh_s serum of known potency is added to each of the tubes. The tubes are shaken and reincubated for one hour. Read for clumping.

Strong clumping in all tubes: **NEGATIVE** for blocking antibodies.

Clumping absent or weak: **POSITIVE** for blocking antibodies to "X" titre.

DEVELOPING TEST OR COOMB'S TEST ON PATIENT'S SERUM

Principle: This test is for a third order of antibodies, or cryptagglutinins. These do not react in saline or bovine albumin and can be demonstrated only with the anti-human globulin or developing serum.

Procedure: When the saline agglutination test is negative, the cells of the first one or two tubes are washed three times in saline to remove unadsorbed globulins, and then resuspended in saline to make a 2% suspension. One drop of the anti-human globulin serum is added to 1 drop of the suspension of cells, and they are reincubated for one hour at 37° C. Centrifuge at 500 rpm for one minute and read macroscopically for agglutination. Clumping indicates the presence of adsorbed immune globulin on the cells or a third order of incomplete antibodies or cryptagglutinins. When the test is positive, the anti-human globulin serum should be added to the higher dilutions of serum in the tubes of the saline agglutination test to learn the titre of these antibodies.

DEVELOPING TEST OR COOMB'S TEST FOR ERYTHROBLASTOSIS

Principle: The cells of a newborn infant are washed with saline to remove all mechanically adhered serum proteins. The anti-human globulin or developing serum is then added. Clumping indicates the presence of a globulin or antibody adsorbed on the red cells. When the test is positive in a newborn infant, it is considered to be diagnostic for erythroblastosis fetalis, even with a subclinical case.

Procedure: The infant's cells are obtained preferably from cord blood, or within the first 24 hours of life. They are suspended in saline and washed with saline three times. The supernatant fluid is then poured off and the cells are made up to a 2% suspension. One drop of the suspension is then mixed with one drop of the anti-human globulin serum and incubated at 37° C. for one hour. Centrifuge at 500 rpm for one minute and read macroscopically for agglutination. Agglutination indicates that the antibodies have been adsorbed *in vivo*.

THE LABORATORY TECHNICIAN IN THE PHYSICIAN'S OFFICE

MRS. HELEN SPICER MENKEL, M.T. (ASCP) *Denver, Colorado*
Laboratory Technician—the physician needs you in his office and you need the experience which may be gained in the physician's office.

Most physicians starting out in private practice feel that a nurse in the office is a **must**, but a laboratory technician is just too expensive a luxury. When the practice gets well on its way, the need for some laboratory work to be done in the office seems a necessity. If the physician knows anything about carrying out laboratory procedures he may attempt to teach his nurse a few simple tests such as albumin and sugar in urine and hemoglobin determinations by the Tallquist method. Many physicians own a microscope and they may teach their nurse to do blood counts also. Perhaps he may have a friend who knows laboratory work or who will be willing to teach his nurse for a few days or evenings. More than likely she has never used a microscope before or perhaps she remembers something about the "scope" from high school or probation days. She is the one who does the doctor's tests, in other words, she is now his laboratory technician.

Today, a boy comes into the office with acute pain in the abdomen and the nurse takes the blood count. The white count is only 5000 per cu. mill. and after so long a time a differential is finished. Only 65 per cent polymorphonuclear. That seems to be quite a normal count. Perhaps the boy doesn't have appendicitis after all. But wait, the nurse hasn't seen all those young "polys" or the occasional metamyelocyte. To an untrained eye they wouldn't mean anything. The doctor is left without the needed information that there is a "marked shift to the left" which knowledge trained laboratory technician would have been able to give him.

Is that skin eruption due to allergy, bacteria or fungus? The doctor puzzles while the nurse stands by. Not so the laboratory technician. She knows just how to fix the slides and cultures to look for bacteria or fungus. She also knows that some skin lesions which show no evidence of organisms may be allergic responses to athletes feet, so scrapings are made from the feet and examined if there is any history of this infection. The laboratory technician scores again! The patient also wins, for the doctor is able to prescribe the right treatment instead of working in the dark or sending the patient to some far off town if he is located away from a medical center.

The returning service man brings up the problem of tropical diseases and the necessity of proper laboratory diagnosis. Stool specimens on these men who come in with vague aches and pains

are quite revealing. The experienced technician is able to recognize intestinal parasites. In a high percentage of these cases, stool specimens contain intestinal parasites. In the last few months, in a small medical practice, four stools examined contained *Endamoeba Histolytica*, three *Endamoeba coli*, one *giardia lamblia* and one contained hookworm ova and the larvae of *Strongyloides*. What a valuable experience for the technician who has been specializing in some large clinical laboratory! Examination for intestinal parasites can be made with little equipment. A simple culture medium can be made by using a Loeffler's blood serum slant with Ringer's solution. If the physician doesn't feel that he wants to invest in an incubator, a small gas stove with a pilot light can always be made to serve your purpose for a home-made incubator. One of the burners from the stove can be removed so that you can attach your rubber tubing for the bunsen burner. This will save installation of special fixtures.

Here is a patient who needs an injection, or some blood taken for serological examination or intravenous. The laboratory technician is just the one who can do this. Hasn't she been doing it all through her training? She is good at getting into the vein instead of over or under or even between the skin!

Assisting with physical examinations is not stressed in our training but what is that to learn after all the other complicated procedures which have been conquered.

Nurses give the metabolism tests in many physician's offices. If the machine works smoothly everything is all right, but one morning the machine just won't work. Now what to do? The doctor fiddles with it and finally it has to go back to the factory or the agent is sent for post haste. The laboratory technician has had experience with different types of machines and knows how to take care of simple breakdowns and probably would be able to straighten out the situation in a hurry.

The colorimeter is expensive and perhaps the doctor doesn't feel that he can purchase this piece of equipment at first but he needs blood sugars and N.P.N.'s done. The technician should be able to take care of that situation. A La Motte type of comparator block and a pair of Cutler Sedimentation tubes will make a fine colorimeter and will check well with your visual colorimeters. Set the standard at 15 or 20 and your unknown the same and if the unknown is stronger dilute with distilled water until the colors match. With these tubes the reading is $S/R \times \text{Factor}$ the same as with the plunger types of colorimeters whereas the other dilution tubes such as the Peebles-Lewis tubes the final calculation is $R/S \times \text{factor}$. If the unknown is less than the standard, dilute the standard and reverse your calculations. This method may seem crude to those who are used to up-to-date equipment

but they can be made to give accurate results and of much value to the small town doctor who needs to know the findings without sending the blood or the patient to some distant hospital laboratory.

These are only a few of the advantages to both physician and technician. Those of us who have worked in large laboratories sometimes fail to take the interest or responsibility we should. We are used to up-to-date equipment and feel that an expensive setup is necessary for accurate work. There is nothing more stimulating to careful, studious work than to spend some time in a physician's office. Here you are close to the patient and see the doctor's orders and prescriptions and are able to observe the results of treatment. It adds to the interest of your work a hundred fold. Here you will find a joy of accomplishment not received in any other type of laboratory. When you have proved your worth to the physician more equipment will soon be coming your way. More physicians might be willing to go to rural communities where they are so sorely needed if they could have the services of well-trained technicians.

In conclusion, I will say, the practicing physician can't afford to be without the services of a well trained technician and most of all, the laboratory technician needs the experience gained in the physician's office.

THE JACK-POT

44 year old colored female admitted in coma. Blood Sugar —1500 mgms. on admission. She received 1025 units of insulin during the 16 hours she lived. Blood Sugar taken just after patient expired was 790 mgms.

Colored male admitted with stabbed heart, apparently lifeless. Blood transfusion started, chest opened and heart sutured. Rapid and uneventful recovery. E.K.G.s throughout convalescence were normal.

32 year old white female admitted with "bulging eyes" B.M.R: Plus 104. Both eyelids were sutured closed, and later an enucleation of the right eye was done. Lugol's and propylthiouracil given. B.M.R. on dismissal was Plus 12. Patient was still rather nervous. Advised to take a rest and build up for thyroidectomy.

Patient, white, female, age 53, with a diagnosis of mylogenous leukemia, was given one X-ray treatment. White count jumped from 850,000 to 1,200,000. Red count was 2,200,000. Ratio of 1:2 instead of 1:700.

—Interesting cases reported by Sr. M. Antonia Klapheke.

THE EDITOR'S PAGE

MORE RECRUITS FOR MEDICAL TECHNOLOGY

"The Oklahoma Society of Medical Technologists," writes Mrs. Dorothy Foreman, their president during 1947-48, "has been working on a project of 'recruiting students for medical technology' for the past year. Miss Mary K. Garrett, MT (ASCP), Chairman of the Public Relations Committee, has assembled information regarding the schools of technology in Oklahoma, along with data regarding educational requirements, scholarships, and answers to the usual questions asked by prospective students. She has prepared a poster and a letter which has been mailed to all high schools in the state (over 450) and to the colleges and universities." Mrs. Lucille Wallace was on the program for the Career Conference held by the University of Oklahoma on April 27, 28, and 29.

In Chicago, the local society of Medical Technologists is a participating organization in "Women's Share in Public Service." This latter organization has as its objectives: "1. To promote greater participation of qualified women in all branches of public service. 2. To improve the political, economic, and social status of all women. 3. To act as a clearing house of information and as a means of joint action and research among the affiliated members." At the fourth annual conference of this organization held on February 21, 1948, the Chicago Society of Medical Technologists was represented by its president, Miss Ruth Feucht, on a panel, "Women in Civil Service."

Mrs. Lavina B. White of Pueblo, Colorado, writes, "We are doing quite a bit with the selling of medical technology to the High School groups; since we have had two 'Occupational Clinics' meetings with the two High Schools here. Lucille Wallace sent me a copy of the booklet from the Registry and it came just before I had to talk to one of the groups. We are having an open house at the C.S. Hospital this month, and expect to invite some of the junior college and H. S. students."

The Committee on Education for the Twin Cities Society of Medical Technologists contacted the colleges in Minnesota which offer a course in Medical Technology. They found the minimum credit hours to be as follows: 3 years of college work (90 semester hours or 135 quarter hours) with 50 semester hours or 62 quarter hours required in the following sciences: Zoology (including histology and parasitology) 12 Semesters or 15 Quarter hours; Chemistry (organic and inorganic required, with a choice of Physiological or Quantitative) 20 Semester or 25 Quarter hours; Bacteriology—4 Semester or 5 Quarter hours; Anatomy or Physiology or Hematology (not all colleges offered all three courses (4 Semester or 5 Quarter hours; Physics (one college considered this unnecessary) 6 Semester or 8 Quarter hours. This, together with the additional year of training in a Clinical

Laboratory under the supervision of a Laboratory Director, i.e., Pathologist or Internist. This course leads to a degree at a baccalaureate level or one may receive a baccalaureate degree in any one of the major fields of Medical Technology and complete the course by taking one year of training in a Clinical Laboratory under the supervision of a Laboratory Director. The committee, Natalie Cremer, Betty Stevenson, Violet Moore, Frances Casey, and Ellen Omernick, announced the results of their studies in the local leaflet, the LENS.

The Harris County Society of Medical Technologists has been invited to become a member organization of the "Occupational Planning Committee" of Houston, Texas. This is an organization composed of civic, educational, business, industrial, and professional groups throughout the city and is for the purpose of coordinating vocational guidance needs. As a participant in such an organization our society will have the opportunity of setting before the community reliable occupational information relating to medical technology.

**WHAT ARE YOU DOING TOWARD RECRUITING
MEDICAL TECHNOLOGISTS?**

ABSTRACTS

AN ALL-GLASS LABORATORY STILL. G. E. Mallory and R. F. Love. Alcohol Tax unit, Bureau of Internal Revenue, San Francisco, Calif. Anal. Chem. 20:94, (Jan) 1948.

These contributors state: "The ordinary still consisting of boiling flask, connecting tube, and condenser, joined by rubber stoppers has the disadvantage that the rubber may be affected by vapors produced in the distillation process. Such action may cause contamination of the distillate and also it hardens and shrinks the rubber so that tight connections are not always maintained and vapor may be lost through leakage. To obviate these difficulties, all glass stills have been devised in which the two rubber stoppers have been replaced by glass joints."

The authors describe and illustrate the apparatus in use and give directions for cleaning the still after use.

SYMPOSIUM ON THE TEACHING OF ELECTRON MICROSCOPY. Anal. Chem. 20:92, (Jan) 1948.

This report includes opinions of outstanding authorities on the subject. S. C. Ellis, of the RCA Laboratories, Princeton, N. J., stated that in his opinion there should be no specialized training to prepare a man solely for electron microscopy, but that a good general education in science is necessary with good training in physics. Those persons taking a course in electron microscopy should have practical experience in operating the microscope and should be able to use it as an aid to research. Electron Microscopy courses are given at the University of Toronto on a graduate level and include the following:

Electron Optics 25 hours. Integrated with general physics. Lenses, aberrations, tolerances, description of the microscope.

Electron Microscopy and Diffraction 25 hours.

A. Microscopy

1. Specimen preparation.
2. Actual Use of the Microscope.
3. Interpretation of results.

B. Diffraction

1. Laue Pattern.
2. Particle Size Determination
3. Uses of Electron Diffraction, Limitations, etc.

Research. The use of the electron microscope in a special research problem. Dr. Ellis emphasized the fact that the best training for research is actual research, and that the electron microscope is not used for research work but to observe the results of research.

The Massachusetts Institute of Technology, The University of Michigan and the National Bureau of Standards offer courses in electron microscopy on the graduate level.

At present electron microscopists are not considered as a spe-

cial group—they are chemists, physicists, or biologists who are using the instrument as a research tool.

THE MICROCOLORIMETRIC DETERMINATION OF SODIUM IN HUMAN BIOLOGIC FLUIDS. Albanese, Anthony A., Ph.D., and Marilyn Lein. Department Pediatrics, New York University College of Medicine and the Children's Medical Service, Bellevue Hospital, New York City. *Jour Lab and Clin Med.* 33:246, (Feb) 1948.

A rapid microcolorimetric procedure for the estimation of sodium content of urine, cerebro-spinal fluid, and various fractions using 0.2 ml samples is described.

A CHEMICAL METHOD OF ESTIMATION OF NICOTINIC ACID IN URINE IN THE PRESENCE OF SUGAR. By Banerjee, Sachchidananda, Naresh Chandra Ghosh, and Gangagobinda, Bhattacharya. (From the Department of Mitra Research in Diabetes, Calcutta School of Tropical Medicine, Calcutta, India) *Jour Bio Chem.* 172:495, (Feb.) 1948.

Because it was reported that intravenous injection of nicotine acid produces hypoglycemia in normal individuals and that a definite improvement in the carbohydrate tolerance of diabetic patients took place after the administration of nicotinic acid, the authors therefore undertook the study of nicotinic acid excretion in diabetic patients. A chemical method for the estimation of nicotinic acid in urine in the presence of sugar is described. Sugar is removed by treating with permanganate and manganese is removed as phosphate and the sugar-free urine is then digested with alkali. Permanganate decolorization is again used and the solution neutralized. Phosphate buffer solution is added and then a suitable aliquot is treated with alcoholic aniline and a solution of cyanogen bromide. A standard nicotinic acid solution is prepared and the unknown compared against it.

The normal daily output of nicotinic acid in the urine is between 1.4 and 5.3 mg%.

AMOEBIASIS, DIAGNOSTIC AND THERAPEUTIC CONSIDERATIONS. By W. J. Alkan M.D., D.T.M.&H (Jerusalem, Palestine. *Jour Trop Med and hyg.* 50:175, (Sept) 1948.

The author suggests the following method of stool examination as more or less fool proof: "Three fresh, warm stool specimens, free from urine and disinfectants, are to be examined. If no *E. histolytica* has been found, the parasite may be recovered from one of three stools passed after a saturated solution of 1 oz. magnesium sulphate crystals has been given in the morning before breakfast." If no amoeba are found, a concentration method should be employed for detecting cysts.

NEWS AND ANNOUNCEMENTS

The American Society of Medical Technologists

SIXTEENTH ANNUAL CONVENTION

Headquarters: Hotel St. Paul, St. Paul, Minnesota

June 7, 8, and 9, 1948

General Chairman: Miss Frieda H. Claussen, 469 Laurel Ave., St. Paul 2

PROGRAM

Sunday: June 6: 1:00 P.M.—Registration opens.

Monday: June 7: 8:30 A.M.—Registration.

Technical and Scientific Exhibits open.

9:00-10:00 A.M.—Continental Room: Presiding: Sr. M. Alcuin, MT (ASCP); Alternate: Mrs. Carl Strolberg, MT (ASCP)

National Anthem.

Invocation: His Excellency, The Most Reverend James J. Byrne, S.T.D., Auxiliary Bishop of St. Paul.

Welcome: The Honorable Luther W. Youngdahl, Governor of the State of Minnesota.

Welcome: Mayor of St. Paul.

Greetings: A. H. Wells, M.D., Board of Registry of Medical Technologists.

Response: Mrs. Lucille Wallace, MT (ASCP), President of the American Society of Medical Technologists.

Announcements: Miss Frieda Claussen, MT (ASCP).

10:00-11:00 A.M.—Panel Discussion: "The Current Field of Medical Technical Laboratory Contributions to Diagnosis and Treatment in Terms of Disturbed Liver Physiology."

E. L. Tuohy, M.D., FACP, Duluth Clinic, Duluth, Minnesota, Panel Coordinator:

"A Merging of Efforts and Understanding by Doctors Ordering Clinical Laboratory Procedures and Medical Technologists Performing Them."

F. J. Hirschboeck, M.D., Duluth Clinic: "A Statement from an Analysis of the Internist's Problems."

Violet Hawkinson, B.S., Junior Scientist, and Margaret Giebenhain, B.S., Department of Internal Medicine, University of Minnesota, Minneapolis, Minn. "Procedures and Problems Involved in Performing Tests for Bile Pigments and Liver Function in General."

Elizabeth G. Frame, Ph.D., Department of Physiological Chemistry, University of Minnesota Medical School, Minneapolis, Minn. "Correlation of Some Liver Function Tests with Serum Protein Fractions."

Section 1: 11:00-11:30—"Special Staining Technics": Mary Frances Gridley, MT (ASCP), U. S. Army Institute of Pathology, Washington, D.C. Paper discussed by T/Sgt. Evelyn Ballou, MT (ASCP).

Section 2: Casino Room: Presiding: Ida Reilly, MT (ASCP).

Alternate: Esther Willbrecht, MT (ASCP).

11:00-11:30—"The Medical Technologist in the Field of Veterinary Science," Agnes Hilden, MT (ASCP), Dept. of Veterinary Science, University of Nevada, Reno, Nevada.

11:30 Both Sections: Visit Exhibits.

1:00-2:00 P.M.—Continental Room: Presiding: Mollie Hill, MT (ASCP); Alternate: Reva Levin, MT (ASCP).

Panel Discussion: *"The Training and Problems of Medical Technologists":

Lall G. Montgomery, M.D., Chairman of Board of Registry of Medical Technologists, Ball Memorial Hospital, Muncie, Indiana, Panel Coordinator: "Responsibilities of Registered Medical Technologists."

Gerald T. Evans, M.D., Director of Course in Medical Technology, University of Minnesota Medical School, Minneapolis, Minnesota: "Guaranteeing the Reliability of Chemical Determinations in Clinical Laboratories."

Rachel Lehman, MT (ASCP), Instructor of Medical Technologists, Indiana University Medical Center, Indianapolis, Indiana: "Simplified Abstract System."

Ruth Church, MT (ASCP); Wilmington, Delaware: "Hiring a Medical Technologist just out of Training School."

Section 1: 2:00-2:30—"Dry Celloidin Technique as Adapted to the Preparation of Eye Tissue," T/Sgt. Evelyn Ballou, MT (ASCP), U. S. Army Institute of Pathology, Washington, D. C.

2:30-3:00—Visit Exhibits.

3:00-3:30—"Radioactive Isotopes," Reverend Sister Mary Antonia, MT (ASCP), Georgetown University Hospital, Washington, D. C.

Paper discussed by Mollie Hill, MT (ASCP), Washington, D. C.

3:30-4:00—"Positive Cephalin-Cholesterol Flocculation Test in X-Ray Workers," Mrs. Nell Butler, MT (ASCP), and James F. Crenshaw, M.D., Seale Harris Clinic, Birmingham, Alabama.

Discussed by Mrs. Hazel Suessenguth, MT (ASCP), Cleveland, Ohio.

4:00-4:30—"Semen Analysis from the Angle of the Clinical Laboratory," Edith Damgaard, MT (ASCP), Duluth Clinic, Duluth, Minnesota.

Discussion by H. J. Walder, M.D., Duluth Clinic, Duluth, Minnesota.

4:30—Visit Exhibits.

—See Entertainment Program.

Section 2: Casino Room: Presiding: Mary Eichman, MT (ASCP); Alternate: Henrietta Lyle, MT (ASCP).

2:00-2:30: "The Hematological Effects of Nitrogen Mustards," Mrs. Edna K. Marks, MT (ASCP), Argonne National Laboratory, University of Chicago, Chicago, Illinois.

2:30-3:00:—"Methods of Washing Glassware," Bernice Elliott, MT (ASCP), Omaha, Nebraska.

3:00-3:30—Visit Exhibits.

3:30-4:00—"Gastric Aspiration Technique, a Guide to Obtaining Specimens for Bacteriological Study," Sylvia Anderson, MT (ASCP), Mt. Sinai Hospital, Milwaukee, Wisconsin.

4:00-4:30—"Equine Encephalomyelitis," Lucille Godelfer, MT (ASCP), Division of Laboratories, Louisiana State Dept. of Health, Central Laboratory, New Orleans, Louisiana.

4:30-4:45—"Can A.S.M.T. Continue to Stand Alone?—UNESCO, WHO?"* Reverend Sister M. Alcuin, MT (ASCP), and 8 students in Medical Technology of the College of St. Scholastica, Duluth, Minnesota:

Paper to be read by Marjorie Cahoon and discussed by Sr. M. Alcuin.

4:45—Visit Exhibits.

See Entertainment Program.

Tuesday: June 8: 9:00-11:30 A.M.—Casino Room: HOUSE OF DELEGATES MEETING.

Section 2: Continental Room: Presiding: Catherine Arens, MT (ASCP); Alternate: Irma Swanson, MT (ASCP).

9:00-9:30—Visit Exhibits.

9:30-10:00—"Some Recent Observations in the Serology of Syphilis," Mrs. Margaret Zwally, M.A., USPHS, Staten Island, New York.

10:00-10:30—"Recent Advances in the Analysis of Vitamins," M. O. Schultze, Ph.D., Professor of Agricultural Biochemistry, University of Minnesota, Agricultural Farm, St. Paul, Minnesota.

10:30-11:00—"Enterobius vermicularis, Human Pinworm," Dorothy J. Hitchcock, MT (ASCP), Department of Bacteriology and Public Health, Michigan State College, East Lansing, Michigan.

11:00-11:30—"Notes on the Preparation of Wright's Stain," John Fitzgerald, MT (ASCP), Mercy Hospital, Portland, Me.

11:30—Visit Exhibits.

12:15—See Entertainment Program.

Section 1—Continental Room: Presiding: Martha Lee, MT (ASCP).

Alternate: Bernice Elliott, MT (ASCP).

2:00-3:00—Panel Discussion: ***"Alpha Delta Theta Invites You": Mary Grieser, MT (ASCP), St. Mary's Hospital, Duluth, Minnesota, Chairman.

Mrs. Fritz Brei, MT (ASCP), Wauwatosa, Wisconsin: "History and Standards of Alpha Delta Theta."

Miss Marian Dallman, President of the Alpha Delta Theta, Minneapolis, Minnesota: "Activities of Alpha Delta Theta—National and Local."

Informal Discussion: "The Relationship of Alpha Delta Theta to the Profession of Medical Technology."

3:00-3:30—"Effect of Total Body X-Irradiation and P_{32} on Peripheral Blood of Normal and Polycythemic Rats," Ellen Skirmont, MT (ASCP), Argonne National Laboratory, University of Chicago, Chicago, Illinois.

3:30-4:30—"The Role of Fungi in Inhalant Allergy," Reva Levin, MT (ASCP), Dr. Leon Unger's Laboratory, Chicago, Illinois.

4:00-4:30—Visit Exhibits.

4:30-5:00—"The Value of the Sedimentation Rate in Diagnosis of Acute Appendicitis," Jane Maghan, MT (ASCP), St. Luke's Hospital, Duluth, Minnesota.

Discussed by A. H. Wells, M.D., Pathologist, St. Luke's Hospital, Duluth, Minn.

Section 2—Casino Room: Presiding: Mary Nix, MT (ASCP). Alternate: Lucille Godelfer, MT (ASCP).

2:00-2:30—"The Role of the Technologist in the Diagnosis of Uterine Cancer," Reverend Sister Mary Norbert, MT (ASCP), Our Lady of Mercy Hospital, Cincinnati, Ohio.

2:30-3:00—"Clinical and Serological Evaluation of 27, 103 Consecutive Slide Tests with Cardioplin Lecithin Antigen and Kline Antigen," B. Levine, M.D., B. S. Kline, M.D., and Suessenguth, MT (ASCP). Read and discussed by Mrs. H. Suessenguth, Mt. Sinai Hospital, Cleveland, Ohio.

3:00-3:30—"A Community Blood Center," Mary Jeanne Reed, MT (ASCP), Junior League Blood Center, Milwaukee, Wisconsin.

3:30-4:00—Visit Exhibits.

4:00-4:30—"The Medical Technologist in Industry," Mrs. Eileen Smith, MT (ASCP), Chemical Analyst, Research Laboratory, General Mills, Inc., Minneapolis, Minnesota.

4:30-5:00—"Liver Function Studies in Chronic Alcoholism Reported on 300 Consecutive Admissions," Marjorie Moss, MT (ASCP), Shadel Sanitarium, Inc., Seattle, Washington.

5:00-5:30—"An Explanation of the Terminology and Definitions Recommended by the Committee for Clarification of the Nomenclature of Cells and Diseases of the Blood and Blood-Forming Organs," Presented through the courtesy of the American Society of Clinical Pathologists, by Margaret E. Hughes, MT (ASCCP), Division of Experimental Medicine, University of Oregon Medical School, Portland, Oregon.

5:30—Visit Exhibits.

Tuesday Evening—Continental Room: Presiding: Oscar Stewart, MT (ASCP).

Alternate: Mrs. Nell Butler, MT (ASCP).

7:30-8:00—"Antibodies Brought Up to Date," Israel Davidsohn, M.D., Director, Department of Pathology, Mt. Sinai Hospital, Chicago, Illinois.

8:00-8:30—"Parasite Problems in Veterans," F. G. Wallace, Ph.D., Veterans' Hospital, Ft. Snelling, St. Paul, Minnesota. (Demonstration).

8:30-9:00—"Determination of Renal Blood Flow and Glomerular Filtration with Para-amino-hippurate and Mannitol," Ben I. Heller, M.D., Veterans' Hospital, Ft. Snelling, St. Paul, Minnesota. (Demonstration).

9:00—Movie: "Operations Crossroads" *Through the courtesy of the Atomic Energy Commission (Prepared by Joint Task Force T of the Army and Navy)—a story in color of the Bikini Scientific Test of the Atom Bomb.

Wednesday—June 9: Tour of the Mayo Clinic, Rochester, Minnesota.

7:30 A.M.—Leave St. Paul for Rochester.

10:00 A.M.—Arrive Rochester—Park at Central School.

10:30 A.M.—Tour Museums of Medical Science.

11:30 A.M.—Leave Museums for Mayo Clinic Building.

12:00 M.—Begin Tour of Mayor Clinic Building.

1:30 P.M.—Luncheon at Central School Cafeteria.***

Informal program featuring A. H. Sanford, M.D., and T. B. McGath, M.D., of the Mayo Clinic Pathology Staff.

3:00 P.M.—Return to St. Paul.

7:00 P.M.—See Entertainment Program.

PROGRAM COMMITTEE

Sr. M. Alcuin, OSB, College of St. Scholastica, Duluth, Chairman

Miss Mollie Hill, 2325 37th St. N.W., Washington 7, D. C.

Miss Martha A. Lee, 14239 Victory Blvd., Van Nuys, California

Mrs. Martha Strolberg, Howard Lake, Minn.

Miss Esther Wilbrecht, New Ulm, Minn.

* Of special interest to students in medical technology.

** Of special interest to students and supervisors.

*** Compliments of the Board of Governors of Mayo Clinic.

SEMINAR IN HEMATOLOGY

University of Minnesota Continuation Center. June 10, 11, 1948

Proposed Program**Thursday, June 10**

- 8:30- 9:00—Complete registration, Dormitory Desk, Center for Continuation Study.
- 9:00- 9:10—Introduction.....George N. Aagaard
- 9:00-10:00—Hemorrhagic diseases—technical aspects
Armand J. Quick.
- 10:15-11:15—Hemorrhagic diseases—clinical application
Armand J. Quick.
- 11:30-12:00—Discussion.
- 12:00—Luncheon in the Center dining room.
- 1:00- 3:00—Erythroblastic anemias with particular relation to the Rh factor—hematology and laboratory aspects
I. Davidsohn.
- 3:10- 3:30—Discussion.

Friday, June 11

- 8:00—Breakfast in the Center dining room.
- 9:00- 9:50—Present therapeutic agents in the treatment of diseases of the blood forming organs.....Howard L. Horns.
- 10:00-10:20—Discussion.
- 10:30-11:20—Megaloblastic anemias of infancy
Edward N. Nelson.
- 11:30-11:50—Discussion.
- 12:00—Luncheon in the Center dining room.
- 1:30- 2:20—Bone marrow technique and morphology
Emil Schleicher.
- 2:30- 3:20—Bone marrow as a diagnostic aid in clinical pathologyA. J. Hertzog.
- 3:30- 4:20—Brucellosis of the bone marrow, Dorothy Sundberg.

FACULTY

George N. Aagaard, Director of Postgraduate Medical Education.
Israel Davidsohn, Director Department of Pathology, Mount Sinai Hospital, Chicago.
A. J. Hertzog, Pathologist, Northwestern Hospital.
Howard L. Horns, Clinical Instructor in Medicine.
Edward N. Nelson, Research Associate in Pediatrics.
Armand J. Quick, Professor of Biochemistry, Marquette University School of Medicine.
Emil M. Schleicher, Hematologist, Minneapolis General Hospital.
Dorothy Sundberg, Assistant Professor of Anatomy.

SPECIAL COMMITTEE ON HEMATOLOGY SYMPOSIUM

Miss Barbara Tucker, Northwestern Hospital Laboratory, Minneapolis,
Chairman
Miss Grace Mary Ederer, Northwestern Hospital Laboratory,
Minneapolis

THE ENTERTAINMENT PROGRAM**Sunday, June 6**

3:30 P.M. to 5:30 P.M.—Sight-seeing bus tour of the Twin Cities, taking in many of the places of interest in St. Paul and Minneapolis. (This tour has already been briefly described in the March issue of the JOURNAL. It may be your only chance to "get around the Twin Towns".)

7:00 to 9:00 P.M.—Informal Reception in the Windsor Room of the St. Paul Hotel. Hostesses: The Minnesota Society of Medical Technologists.

Monday, June 7

6:30 P.M.—Smorgasbord at White Pine Inn, Bayport, Minn. Scandinavian dancers in native costume will provide entertainment. This will be followed by a talk on "The Evolution of Medical Technology," by Dr. Kano Ikeda.

Busses will leave the St. Paul Hotel for White Pine Inn at 5:30 P.M.

Tuesday, June 8

12:15 P.M.—Luncheon in the Continental Room, St. Paul Hotel. A Style Show will be presented by the Field-Schlick Department Store. The guest speaker will be Dr. Lall Montgomery, Chairman of the Board of Registry.

Wednesday, June 9

7:00 P.M.—Annual Banquet, Continental Room, St. Paul Hotel. The announcement of the awards for the best papers and exhibits will be made by Dr. Kano Ikeda. Several events of unusual interest are being planned to make this an entertaining evening.

(NOTE: Although the Banquet is, by tradition, a formal affair, informal attire will be entirely acceptable. There are many who prefer it so, and we want EVERYBODY to come.)

(NOTE: Plans for the entertainment of visiting Sisters are underway. Since these have not been completed at this time, they will be announced at the convention.)

ENTERTAINMENT COMMITTEE

Miss Loretta Laughlin, 315 No. 11th St., Benson, Minn., Chairman

Miss Dorothy Misjuk, Minneapolis General Hospital, Laboratory,
Minneapolis, Co-Chairman

SUB-COMMITTEES**INFORMAL RECEPTION SUNDAY EVENING**

Miss Constance Olson, Miss Anne Dimunation of the Northwestern Hospital Laboratory, Minneapolis

SMORGASBORD, MONDAY EVENING

Miss Ellen Omernick, Miss Dorothy Misjuk of the Minneapolis General Hospital Laboratory, Minneapolis

ST. PAUL HOTEL LUNCHEON, TUESDAY NOON

Miss Elizabeth Schneider, Miss Doris Hansen, Miss Audrey Gutterson
of the Ancker Hospital Laboratory, St. Paul 2

BANQUET, WEDNESDAY EVENING

Miss Marjorie Due, Miss Elizabeth Ann Stuber, Miss Marian Cody,
Mrs. Nabuko Mizuno, of the Charles T. Miller Hospital Laboratory,
St. Paul 2

TRANSPORTATION

Miss Frances Casey, 1466 Midway Parkway, St. Paul 4, Chairman
Miss Katherine Hartwell, 1446 Fairmount Ave., St. Paul
Miss Marguerite Lutgen, Ancker Hospital Laboratory, St. Paul 1
Miss Evelyn Hultkrans, 1975 Berkley Ave., St. Paul 5
Miss Magdalene Dege, 1141 Fauquier St., St. Paul
Miss Margaret Bunde, 511 Sherburne Ave., St. Paul

SPEAKERS' SUPPLIES

Miss Mary Conroy, 865 Iglehart Ave., St. Paul 4, Chairman
Mr. Chauncey Winbigler, St. Joseph's Hospital Laboratory, St. Paul 2
Miss Dorothy Magaw, Wilder Dispensary Laboratory, St. Paul 2

SIGN COMMITTEE

Mrs. Virginia Bray Paterson, 420 University Ave. S. E., Minneapolis,
Chairman
Mrs. Vivian Wolff, St. Peter Hospital, St. Peter, Minn.
Miss Donna Luker, 3724 Pillsbury Ave., Minneapolis

COMMITTEE ON STUDENT RELATIONS

Miss Mary Grieser, 1831 Wallace Ave., Duluth, Chairman
Miss Marian Dallman, 330 11th Ave., S. E., Minneapolis
Mrs. Fritz Brei, 8435 Kenyon Ave., Wauwatosa, Wisconsin
Miss Ruth Feaster, 2453 Garfield Ave. South, Minneapolis 5, (Housing)

REGISTRATION

Miss Margaret Strane, Miller Hospital Laboratory, St. Paul 2, Chairman
Miss Eleanor Eggleston, 3407 15th Ave., So. Minneapolis 7
Miss Edith Damgaard, Duluth Clinic, Duluth
Miss Marion Polasky, 315 No. 61st Ave. W., Duluth 7
Miss Lucille Peterson, Children's Hospital Laboratory, St. Paul 2
Miss Irma Swanson, Abbott Hospital Laboratory, Minneapolis 4
Mrs. Virginia Rask, 143 E. Arion St., St. Paul 7
Miss Sigrid Serum, Minneapolis General Hospital Laboratory,
Minneapolis
Miss Marjorie Ellison, Minneapolis General Hospital Laboratory,
Minneapolis
Miss Wilma Hayes, Asbury Hospital, Minneapolis

ROCHESTER TOUR COMMITTEE

Miss Elizabeth Maclay, 1141 9½ Ave. S.E. Rochester, Minn., Chairman
Arlene Magnusson, Mayo Clinic Laboratories, Rochester
(Printing & Signs)
Patricia Hass, Mayo Clinic Laboratories, Rochester
(Menus & Decorations)
Helen Cook, Mayo Clinic Laboratories, Rochester (Luncheon)
Margaret Dunnette, Mayo Clinic Laboratories, Rochester (Program)
Doug Williams, Mayo Clinic Laboratories, Rochester
(Director of Tour in Charge of General Service)

TECHNICAL EXHIBITS

- Miss Cecelia Kortuem, 1164 No. Dearborn, Chicago 10
- Miss Donna Keller, 1349 W. Idaho, St. Paul 4
- Miss Marion Bartl, Swedish Hospital Laboratory, Minneapolis

SCIENTIFIC EXHIBITS

- Miss Grace Mary Ederer, Northwestern Hospital Laboratory,
Minneapolis, Chairman
- Mrs. Agatha Caylor, Litchfield, Minn.
- Miss June Tyson, St. Joseph's Hospital, Brainerd
- Sister M. Michael Kilmer, St. Cloud Hospital, St. Cloud

RECEPTION

- Miss Patricia Bolger, 724 Fourth St. South, Moorhead, Minn., Chairman
- Miss Jane Maghan, 125 E. Linden St., Duluth
- Mrs. Delores Richter, 836 Lowry Bldg., St. Paul 2
- Miss Marie Dammann, 1210 Lowry Medical Arts Bldg., St. Paul 2
- Sister M. Emerita, St. Gabriel's Hospital, Little Falls
- Miss Frances Anderson, Fairview Hospital Laboratory, Minneapolis
- Miss Marian J. Anderson, 1765 Laurel Ave., St. Paul
- Miss Thelma Erickson, Ancker Hospital Laboratory, St. Paul 2
- Miss Jeanne Traxler, St. Joseph's Hospital Laboratory, St. Paul 2
- Miss Ruth Cardinal, St. Joseph's Hospital Laboratory, St. Paul, 2
- Mrs. Shirley Gitis, Bethesda Hospital Laboratory, St. Paul 1
- Miss Dorothy Jane Johnson, St. Luke's Hospital Laboratory, St. Paul 2

SISTERS' HOSPITALITY AND RESERVATIONS

- Sister Marcella Marie Pavlik, St. Joseph's Hospital, St. Paul 2, Chairman
- Sister M. Conchessa Burbridge, St. Mary's Hospital, Minneapolis
- Miss Jeanne Traxler, St. Joseph's Hospital, St. Paul 2
- Miss Ruth Cardinal, St. Joseph's Hospital, St. Paul 2

COMMITTEE ON AWARDS

- Miss Grace Ballard, 925 No. 13th St., Milwaukee 3, Wis., Chairman
- Dr. Kano Ikeda, Miller Hospital Laboratory, St. Paul 2, Minn.
- Dr. J. A. Nelson, 108 West 6th St., Tulsa, Oklahoma
- Dr. Norbert Enzer, 908 No. 12th St., Milwaukee 3, Wisconsin
- Miss Catherine Hanitch, Oak Terrace, Minn.
- Mr. Homer L. Spencer, 411 Medical Arts Bldg., Tulsa, Oklahoma

PUBLICITY

- Mrs. Eileen Smith, 815 Kenwood Parkway, Minneapolis 5, Chairman
- Mrs. Dorothy Hand, 1210 Lowry Medical Arts Bldg., St. Paul 2
- Miss Shirley Finucane, 2712 Emerson Ave., So. Minneapolis
- Mrs. Dorothy Schommer, 1497 W. Idaho, St. Paul 4

SCIENTIFIC EXHIBITS

ASMT National Convention

June 7, 8, and 9, 1948

1. Junior League Blood Center of Milwaukee—Wisconsin Medical Technologists.
2. *Enterobius vermicularis*—Dorothy J. Hitchcock, Instructor in Parasitology, Michigan State College, East Lansing, Mich.
3. Miniature St. John's Hospital—Sister Roselda Mercier, St. John's Hospital, Springfield, Illinois.
4. Role of Fungi in Inhalent Allergy—Reva Levin and Dr. Leon Unger, Chicago, Illinois.
5. "The New Look in the Laboratory"—District Number 3, Minnesota.
6. Modifications of Laboratory Equipment — University of Minnesota, Minneapolis.
7. Unusual Urinary Cellular Elements Related to Heavy Metal Therapy—Mary Frances James, University of Alabama, Birmingham.
8. Methods of Washing Laboratory Glassware—Bernice Elliott, Omaha, Nebraska.
9. A Map of the Illinois District of Medical Technology—Illinois Medical Technologists.
10. "Educational Exhibit"—The American Society of X-Ray Technicians.
11. Demonstration of Commonly Used Special Stains—Melvin Schadewald, M.S., Ph.D., Veterans Administration Hospital Minneapolis, Minn.
12. The Board of Registry Exhibit — Board of Registry of ASCP, Muncie, Indiana.
13. The American Society of Medical Technologists Exhibit.

All exhibits should be at the following address by June 1, 1948:

ASMT Scientific Exhibits St. Paul Hotel, St. Paul, Minnesota.

Technical Exhibits in addition to those listed in March Journal:
Lederle Laboratories (Division Am. Cyanamid Co.) New York;
C. F. Anderson & Co., Inc., Minneapolis, Minn.

Respectfully submitted,

GRACE MARY EDERER, M. T.
Scientific Exhibit Chairman,
Northwestern Hospital, Minneapolis, Minn.

NOTICE

There are a number of "labels" advertising the ASMT Convention available to affiliated societies and individual members. Cost—\$1.00 per hundred or \$5.00 per thousand. Use—personal mail, laboratory and hospital supply orders, Official Publications and notices, place cards and favors. Send orders to Mrs. Eileen B. Smith, Publicity Chairman, 815 Kenwood Parkway, Minneapolis, Minnesota.

COMMITTEE ON STANDARDS AND STUDIES

The Standards and Studies Committee is mailing out a questionnaire. This is being sent to A.M.A. approved hospitals, and all clinics and doctors' offices employing technicians, whose addresses we have. If you know of other clinics and offices employing technicians, please notify any member of the committee, so that a questionnaire may be mailed to them.

As Chairman, I want to take this opportunity to thank the committee for their untiring efforts. I also wish to thank Sister Mary Antonia (Georgetown Univ. Hospital, Washington, D. C.) and Mrs. Winogene McIntyre, Washington, D. C. These people have put in countless hours of effort and labor as a courtesy.

If our returns are received on time, we shall give you a complete analysis and evaluation at the convention in St. Paul.

MOLLIE L. HILL, M.T. (ASCP),
Chairman, Standards and Studies Com.

ALPHA MU TAU FRATERNITY

Mr. George Kneeland, MT (ASCP), 950 E. 59th St., Chicago 37, Illinois, Secretary-Treasurer of Alpha Mu Tau (National Medical Technology Society) announces that this organization was chartered February 20, 1948, in the state of Illinois. Its objective is the professional advancement of medical technology. Membership is limited to registered Medical Technologists who are also in good standing with the American Society of Medical Technologists. Charter members are those who have in some way contributed to the advancement of medical technology or who have by their long service in the profession demonstrated their interest and ability. After June 9, 1948, membership will be closed to all except past national presidents and Honorary members.

LOCAL AND STATE SOCIETIES

- ALABAMA: Miss Lucille Godelfer, 2928 Bell St., New Orleans, La.
- ARIZONA: Miss Mary Wood, Yakima Medical & Surgical Clinic, Yakima, Washington.
- ARKANSAS: Miss Lila L. Church, 2116 Orange St., North Little Rock.
- CALIFORNIA: Miss Jeanne Jorgenson, 900 Modoc St., Berkeley 7, Cal., or Miss Genevieve Walters, 1204 Ozone St., Santa Monica, Sec'y C.S.M.T. Convention on May 7, 8 at Huntington Mem. Hosp., Pasadena.
- COLORADO: Miss Pauline Kurachi, Colorado State Hospital, Pueblo, Sec'y., Colorado S.M.T.
- CONNECTICUT: Acting President: Miss Florence Pease.
Acting Sec'y-Treas.: Miss Lydia Brownhill, Meriden Hospital, Meriden.
- DELAWARE: President: Miss Ruth M. Church, Wilmington General Hospital, Wilmington.
President-elect: Miss Georgene Withers, Del. Hosp., Wilmington.
Sec'y-Treas.: Miss Sarah N. Bruce, Mem. Hosp., Wilmington.
Directors: Mrs. Marie D. Schreyer, 527 Delaware Ave., New Castle.
Mrs. Evelyn G. Scott, 4 Champlain, Bellemoor.
- DISTRICT of COLUMBIA: Mrs. Mary Agnes Comly, 109 Park St., Rockville, M. D., Sec'y., or Miss Ida Reilly, Roanoke Hospital Association, Roanoke, Va.
- FLORIDA: Miss Lucille Godelfer, 2928 Bell St., New Orleans, La.
- GEORGIA: Miss Lucille Godelfer (see above), or Miss Elizabeth E. Paulson, Sec'y., Savannah S.M.T., 515 East 41st St., Savannah.
- IDAHO: Miss Mary Wood, Yakima Medical & Surgical Clinic, Yakima, Washington.
- ILLINOIS: Miss Helen Gurley, Mt. Sinai Hospital, Chicago, Sec'y., Illinois M.T.s Association, or Miss Edna Luneke, 428 North College, Grand Rapids, Michigan.
- INDIANA: Miss Edna Luneke (see above).
- IOWA: Miss Edna Luneke (see above).
- KANSAS: President: Mary Howard, 827 Porter, Wichita.
Vice-President: Sister M. Florinam, Wichita Hospital, Wichita.
Sec'y-Treas.: Miss Mary Louise Kerschenm, 148 So. Charles, Wichita.
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- LOUISIANA: Miss Lucille Godelfer (see above).
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- MARYLAND: President: Miss Betsy H. Schmitz, 118 Midhurst Road, Baltimore.
Vice-President: Mrs. Florence Singer, 2707 Liberty Heights, Ave., Baltimore.
Corresponding Secretary: Mrs. Norma McElvain, 3068 Tenth St., Baltimore.
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- MICHIGAN: Miss Edna Luneke (see above).
- MINNESOTA: Membership Chairman: Miss Frieda Claussen, 469 Laurel Ave., St. Paul, Minnesota.

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- MONTANA: Miss Mary Wood (see above).
- NEBRASKA: Miss Selma Anderson, University of Nebraska, College of Dentistry, Lincoln 8, Sec'y., Nebraska S.M.T. or Miss Edna Luneke (see above).
- NEVADA: Miss Mary Wood (see above).
- NEW HAMPSHIRE: Miss Annie Clark, Higgins, Hospital, Wolfesboro, Sec'y., N.H.S.M.T., or Miss Elizabeth Frey (see above).
- NEW JERSEY: Miss Ida Reilly (see above).
- NEW MEXICO: President: Sister Charles Miriam Strassell, St. Joseph's Hospital, Albuquerque.
Vice-President: Sister Joan of Arc (Allard), St. Anthony's Hospital, Las Vegas.
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- NORTH DAKOTA: Sister M. Eleanor Mischel, St. Alexius Hospital, Bismarck.
- OHIO: Secretary: Miss Mary Lewis, 151 Storer Ave., Akron 2.
- OKLAHOMA: Membership Chairman: Anne Adwan, 1115 Medical Arts Bldg., Oklahoma City 2.
- OREGON: Miss Agnes Marie Lyman, St. Vincent's Hospital, Portland, Sec'y., Oregon S.M.T., or Miss Mary Wood (see above).
- PENNSYLVANIA: Miss Licia G. Gambescia, 3101 School Lane, Drexel Hill, Sec'y., P.S.M.T. & L.T., or Miss Elizabeth Frey (see above).
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- SOUTH CAROLINA: Miss Ida Reilly (see above).
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Sec'y-Treas.: Sister Mary Mildred, St. Joseph's Hospital, Mitchell.
- TENNESSEE: Miss June M. Brown, 3515 Granny White Road, Nashville, Sec'y., Tenn. S.M.T., or Moss Lucille Godelfer (see above).
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- UTAH: Miss Marguerite George, Holy Cross Hospital, Salt Lake City.
Sec'y., Utah S.M.T., or Miss Mary Wood (see above).
- VERMONT: Miss Elizabeth Frey (see above).
- VIRGINIA: Miss Ida Reilly (see above).

WASHINGTON: Miss Mary Wood (see above).

WEST VIRGINIA: President: Doris E. Boon, Charleston.

Vice-President: Mrs. Lena Huffman, Huntington.

Secretary: Constance Peterkin, Parkersburg.

Treasurer: Dorothy Crawford, St. Albans.

WISCONSIN: Miss Margaret Foley, 2039 N. State St., Milwaukee, Sec'y., Wisc. Ass'n. of M.T.s, or Miss Edna Luneke (see above).

WYOMING: Membership Chairman: Miss Georgia Schmidt, 708 East 21st., Cheyene.

TERRITORIES AND FOREIGN: Miss Jeanne Jorgenson, 900 Modoc St., Berkeley 7, California.

State societies in Connecticut, New Jersey, North Dakota, New Mexico, Rhode Island, South Dakota, West Virginia and Wyoming have held their initial meetings and are applying for affiliation with the American Society of Medical Technologists. If you are a member of A.S.M.T. and have not yet joined one of the above organizations or an affiliated state society, write for an application blank. These may be obtained from the A.S.M.T. Executive Office, Medical Center Bldg., Lafayette, Louisiana, or from the Membership Chairman and state secretaries as listed. American Society Members of the other eleven states should make known their interest in organizing a state society. This can be accomplished through the Membership Counsellor as listed for the respective states.

CURRENT MEETINGS

Washington, D. C. Seminar on May 1, 1948.

Mitchell, South Dakota, organizational meeting, April 8, 1948.

Bismark, North Dakota, organizational meeting, April 11, 1948.

Kansas, Convention, April 23, 1948.

Tulsa, Oklahoma, Convention, April 17 and 18, 1948.

Pasadena, California, May 7 and 8, 1948.

Dallas, Texas, Convention, April 23 and 24, 1948.

Lincoln, Nebraska, Convention, April 24, 1948.

Detroit, Michigan, Convention, April 9, 1948.

Newark, New Jersey, Convention, April 9, 1948.

Albuquerque, New Mexico, organizational meeting, April 18, 1948.

Denver, Colorado, Convention, May 23, 1948.

Buffalo, New York, Seminar, May 8, 1948.

Wilmington, Delaware, Seminar, May 22, 1948.

The dates of your meetings will be published in the Journal in the next issue after we receive that information. Papers presented before these groups are accepted for possible publication. They may be sent to the Editor of A.J.M.T., 2119 Arbor Avenue, Houston 4, Texas.

APPLICATION FOR HOTEL RESERVATION
AMERICAN SOCIETY OF MEDICAL TECHNOLOGISTS
ANNUAL CONVENTION, JUNE 7, 8, 9, 1948
ST. PAUL, MINNESOTA

Please make hotel reservations noted below, and mail to Chairman of Local Arrangements Committee; Miss Frieda H. Claussen, 469 Laurel Ave., St. Paul 2, Minnesota.

The St. Paul Hotel is headquarters hotel. The management will make every effort to house all ASMT members, delegates and visitors at this hotel. There are always, necessarily a limited number of single rooms available in one hotel. In order to accommodate as many as possible in the headquarters hotel, we would appreciate it very much if delegates and members will arrange to share rooms, or express their willingness to be assigned a roommate by our housing bureau. Will one occupant of a room please make out this form and indicate name and address of other occupant(s).

Any overflow of reservations will be accommodated at the Lowry Hotel, one and one-half blocks from headquarters, where the rates are the same.

If, after your reservation has been made, you find that you will be unable to attend the convention, please be sure to cancel your reservation directly with the hotel.

Date of Arrival _____ Time _____

Date of Departure _____ Time _____

Name _____

Address _____

Please put check opposite the accommodations desired.

Room for 1 per day	Room for 2 Double bed	Room for 2 Twin beds	Room to accommodate 3 or 4 Persons	Suite—Living Room Bedroom and Bath
\$4.00	\$5.50	\$6.50	\$3.00 per person	for One \$12.50
4.50	6.00	7.00		15.50
5.00	6.50	7.50	Room to accommodate 5 or 6 Persons	for Two 14.00
5.50	7.00	8.50	\$2.75 per person	17.50
6.00		9.00		
6.50				

If no room is available at the rate requested, reservation will be made at the next higher rate.

All guest rooms in the St. Paul Hotel have outside exposure, yet are free from carline and street noises. Every room is equipped with combination tub and shower.

All Sisters desiring housing accommodations, please communicate with Sister Marcella Marie, Chairman Sisters' Hospitality Committee, St. Joseph's Hospital, St. Paul 2, Minn.

ENTERTAINMENT RESERVATION

	Number of Reservations
Sightseeing Bus Tour around Twin Cities (35 miles) Sunday afternoon, June 6.....	\$.75_____
Informal Reception St. Paul Hotel Sunday evening, June 6.....	No charge_____
Smorgasbord Dinner, White Pine Inn.....	3.00_____
Bus Transportation to White Pine Inn..... Monday evening, June 7	.65_____
Luncheon, St. Paul Hotel (Style Show—Door Prizes) Tuesday Noon, June 8	2.00_____
Luncheon, Rochester, Minnesota.....	No charge_____
Bus Transportation to Rochester and Return (170 mi.) Wednesday, June 9	2.00_____
Formal Banquet, St. Paul Hotel..... Wednesday, evening, June 9	5.00_____
TOTAL	\$13.40

Guests of members are welcome at all entertainment. Rochester reservations limited to 400.

SEMINAR RESERVATION

Please make_____reservation for me for the Haematology Seminar to be held June 10 and 11, 1948 at the University of Minnesota.

Address _____

Name _____

Please check and mail all reservations to Miss Frieda Claussen, 469 Laurel Ave., St. Paul 2, Minn.

